H I L G A R D I A

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# BIONOMICS OF THE WALNUT HUSK FLY, RHAGOLETIS COMPLETA[1,2,3]

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## INTRODUCTION

THE PERSIAN WALNUT (*Juglans regia*) industry of the United States is centered in the state of California, where approximately 97 per cent of the tonnage is produced.[(4)] Batchelor[(4)] estimates that the bearing acreage for 1930 was 95,900 acres, with an expected increase of approximately 6,000 acres a year for the next three years. Concerning the economics of the industry he states, "Pest control is becoming an increasingly costly operation in some localities, and this is causing the replanting of former walnut acreage to crops not subject to the pests in question."

The recorded insect fauna[(3)] of the genus *Juglans* numbers over 300 species, only a small number of which occur in California. Before the advent of the walnut husk fly, *Rhagoletis completa* Cresson, the codling moth, *Carpocapsa pomonella* (Linn.), and the walnut aphid, *Chromaphis juglandicola* (Kalt.) were the only species considered to be of major importance to the industry. With the addition of another major pest, the production costs will necessarily be increased in those localities where susceptible varieties are grown.

It is of interest to note that *Rhagoletis completa* is the first species of Trypetidae of major economic importance to become established in California.

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[2] Paper No. 280, University of California Graduate School of Tropical Agriculture and Citrus Experiment Station, Riverside, California.

[3] Presented as a thesis to the graduate division of the University of California, December, 1932, in partial fulfillment of the requirements for the degree of Doctor of Philosophy.

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The project reported in this paper was undertaken in 1928, when general observations were made on the field biology and economics of the insect. An intensive study of the problem was begun in 1929 and continued through the season of 1932.

## HISTORY

This insect has been definitely known to occur in California since 1926,[5] and it may logically be concluded that it was introduced prior to that time. In October of that year S. E. Flanders found dipterous larvae feeding in the exocarp of several varieties of Persian walnut in the Chino section, San Bernardino County. At that time it was suspected that the insect was *Rhagoletis suavis* (Loew), which occurs in eastern United States. Samples of infested nuts were placed under suitable conditions for the insect to complete its life cycle. When adults emerged from the material, they proved to be *Euxesta putricola* Cole, a species with scavenger habits. It was then assumed that the husks of the infested nuts had been mechanically injured and had been subjected to attacks by scavenger species.

However, during the following season (1927) walnuts in the same grove, as well as in several groves that either adjoined or were in close proximity, were noted to have larvae feeding within their green husks. The circumstances surrounding these observations indicated that the insect concerned had deposited its eggs in normal, healthy husk tissue. Again samples were taken for rearing purposes, from which several species of scavenger flies emerged shortly after collection; and the following June (1928) a species of Trypetidae emerged. Specimens of the latter were forwarded to the United States National Museum, where they were identified as *Rhagoletis juglandis* Cresson. This species was described in 1920[11] from specimens reared from larvae feeding in the exocarp of a variety of *Juglans regia* growing on the property of C. R. Biederman in Carr Canyon, Huachuca Mountains, Arizona. The official identification was accepted at that time without further verification.

Since *Rhagoletis juglandis* was reported to be responsible for injury to Persian walnuts in the type locality, a preliminary study of the insect in California seemed advisable. Observations in 1928 showed that the biology of the insect differed materially from that reported by Biederman.[11] He says, "[the fly] appears toward the end of June. . . . The earlier larvae go to the ground by a silk thread, for pupation, but most of them stay in the hull till the nut falls, and pupate in it." Apparently, therefore, much could be gained by visiting Carr Canyon and studying the insect in the locality from which it was first collected and described.

On August 13, 1929, K. L. Wolff and the author reached the homestead of C. R. Biederman in Carr Canyon. Adults were very abundant at that time, and serious damage was resulting from their activity. That the insect in California was entirely different from the one in Arizona was immediately evident. Detailed observations resulted in information contrary to that reported by Biederman. The larvae did not go to the ground by a silken thread, and only a very small number pupated in the walnut husk. Another trypetid was collected at this time from walnuts in Carr Canyon and subsequent study showed that it had not been described. Specimens of flies from Arizona and from California were forwarded to E. T. Cresson for determination. He described the insect from California as *Rhagoletis suavis completa* n. subsp., and the undescribed species from Arizona as *R. boycei* n. sp. These descriptions were published in December, 1929.[12] Cresson examined nine specimens collected in 1917 and 1918 in Texas, that were in the United States National Museum, and considered them to be conspecific with *R. suavis completa*.

One series of specimens in the National Museum, collected at Pecan Bayou, Texas, in 1918, was incorrectly labeled *Rhagoletis juglandis*. Likewise a single specimen from Manhattan, Kansas, in 1921, was incorrectly determined. Through the coöperation of Professor R. C. Smith, the Kansas material was forwarded to the author at the Philadelphia Academy of Natural Sciences, where it was studied in comparison with available type material of the several species of *Rhagoletis* that attack walnut. There were four specimens in the Kansas material collected at Manhattan in 1920: three of them were labeled *juglandis* and the other *suavis*. These four specimens were incorrectly labeled, for they proved to be *completa*. There were four more specimens of *suavis* correctly determined, collected by F. Marlatt, Riley County, Kansas. The date of collection was not given, though circumstances indicate that it was prior to 1910. Large numbers of specimens collected from walnuts in 1930 in Nebraska, Kansas, and Texas, were studied, and *juglandis* was not found in any of this material. Furthermore, all material from Texas was conspecific with that from California, as was the greater portion of that from Kansas, while the remainder from Kansas was *suavis*. The specimens from Nebraska were consistently smaller than *completa* from other sections, though other morphologic differences were not evident. Detailed studies of the systematics of *suavis* and the accepted subspecies *completa* showed that sufficient differences existed in wing markings, male genitalia, and biology to warrant the elevation of *completa* to species rank.

*Probable Method of Introduction into California.*—In 1930, C. C. Delphey and G. A. Pohl, agricultural inspectors, discovered isolated infestations in wild walnuts at Mountain View, Devore, and Devil's Canyon, San Bernardino County (fig. 1). These infestations are located near the main artery of auto traffic leading into southern California

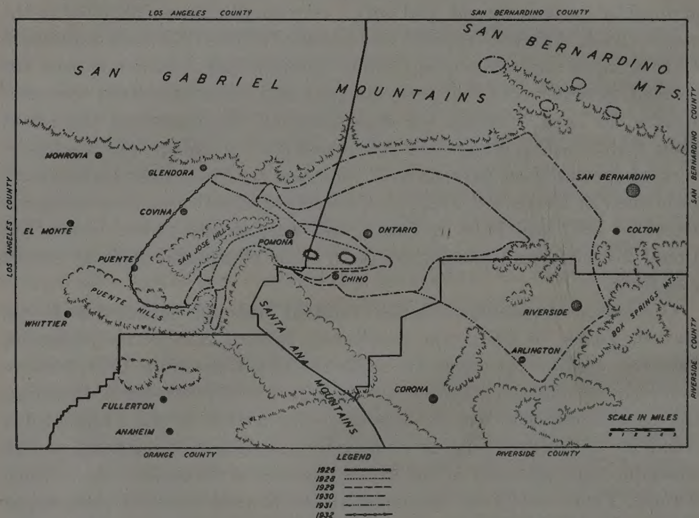



Fig. 1. Distribution of *Rhagoletis completa* in California, showing approximate yearly increase in size of infested area. (Based on survey data obtained from the California State Department of Agriculture and the agricultural commissioners' offices of Los Angeles and San Bernardino counties.)

The rectangle outlined includes only slightly more than that area of each county required to show the extent of infestation. Solid lines within rectangle represent boundary lines of each county.

from eastern states. Therefore it is suspected that auto tourists gathered infested black walnuts when passing through Kansas, Oklahoma, Texas, or New Mexico, and consumed the kernels or otherwise disposed of the walnuts at one of the above locations in California. Since walnuts in the husk have been restricted from entering the state, many lots have been intercepted at the border by the inspecting officers, and in some instances the presence of *Rhagoletis* larvae has been reported. This fact indicates the probable mode of entry.

## TAXONOMY AND TECHNICAL DESCRIPTION OF STAGES

*Adult.—Rhagoletis completa* (figs. 2 and 3) was described in 1929 by E. T. Cresson, Jr., who considered it a subspecies of *suavis.*[12] Regarding the insect he states: "This is no doubt a subspecies of the eastern black walnut maggot, in which the median hyaline costal triangle of *suavis* assumes a more or less complete transverse band, extending at least to the fifth vein." His original description follows:

*Rhagoletis suavis* subsp. *completa* new subspecies.—Of a general ferruginous to tawny color; with a faint median stripe, lateral and posterior portion of the mesonotum, upper portion of pleura including metanotum, forecoxae, all femora beneath and basal portions of second to fifth abdominal segments, brown to black. Frons and occiput medianly, mesonotum in general, pectus, abdomen in general, ferruginous. Face, antennae, palpi, posterior orbits, humeri, notopleural stripe, scutellum, apices of second to fourth abdominal segments, femora above, all tibiae and tarsi, yellowish white. Mesonotum medianly (leaving lateral margins and posterior area including prescutellar bristles, shining) subopaque, ochreous pollinose, with numerous, short, golden hairs. Bristles of posterior orbits pale. Wings as figured . . . . . . ; similar to *suavis*, but the hyaline area beginning at the costa beyond tip of first vein extends as a transverse band, at least to the fifth vein, generally to the inferior margin. In some specimens there is a diluted spot in the portion of the infuscated band in the apical part of the discal cell, sometimes occupying almost entire apical portion with a streak of same dilution extending through the proximo-median band in discal cell. The median hyaline band often broader than is shown in the figure. Length, 4 to 7 mm.

*Type.*—Male; Chino, San Bernardino County, California (A. M. Boyce, August 10, 1928; infesting the exocarp of the Persian walnut, *Juglans regia* Linn.), (A.N.S.P. No. 6341). Paratypes. —3 ♀; topotypical (A.N.S.P.). 3 ♂, 4 ♀; topotypical, August 15, 1925 (U.S.N.M.). 2 ♂, 3 ♀; Pomona, Los Angeles County, California, August 10, 1929 (L. Gammon, California State Department of Agriculture).

Subsequently an examination of hundreds of specimens, which were not available to Cresson when he described *Rhagoletis completa,* showed wide variations in certain color characters as well as in the pattern of infuscated areas on the wing of the type specimen. These variations are briefly recorded as follows: Lateral and posterior portion of the mesonotum, upper portion of pleura including metanotum, fore coxae, all femora, pectus, and venter of abdomen may be totally ferruginous or of darker colors ranging to shining black; antennae, palpi, posterior orbits, apices of second to fourth abdominal segments, all tibiae, and tarsi, most commonly ferruginous to tawny in color though frequently the two distal tarsal joints are brownish; halteres, yellowish-white. Hyaline area of wing that begins at costa beyond tip of first vein (R 1) extends as a transverse band at least to third vein (R 4 + 5) though generally to inferior margin. However, in many instances, a narrow, longitudinal, infuscated area at or below third vein (R 4 + 5) connects the two trans-

Fig. 2. *Rhagoletis completa*, male, dorsal aspect showing yellowish-white scutellum, wing markings, characteristic position of wings, and rounded tip of abdomen.

Fig. 3. *Rhagoletis completa*, male, lateral aspect, showing yellowish-white lateral stripe on the otherwise dark thorax, extended proboscis, claspers, and anus.

verse infuscated bands, in which case a transverse hyaline band extends beyond to inferior margin. The wing marking is the most important character used in identifying this insect.

The elevation of Cresson's subspecies *completa* to species rank was based on the following differences: Normally *suavis* is appreciably larger than *completa*. The second and third infuscated transverse bands on the wing of *suavis* (fig. 4 *B*) are always joined in the basal portion of the

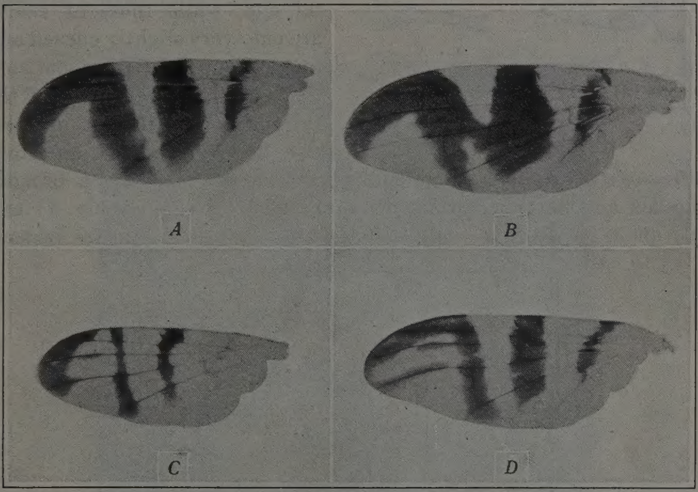



Fig. 4. Left wing of male of the walnut-inhabiting species of *Rhagoletis*, indicating relative size, shape, and pattern of darkened areas. *A*, *R. completa;* *B*, *R. suavis;* *C*, *R. juglandis;* and *D*, *R. boycei.*

first posterior cell (5th R) or in the discal cell (1st M 2). In some instances these two bands are united completely and extend to the inferior margin of the wing as one wide band. However, a relatively narrow infuscated area generally connects these two bands in the discal cell (1st M 2). They separate later and both continue toward the inferior margin. As a result of the union between these two bands a transverse triangular hyaline area is produced, the base of which is a portion of the costal margin, and the apex generally extends at least to the fourth vein (M 1 + 2). It has already been stated that in *completa* the hyaline area transversing the wing between the second and third infuscated bands generally extends to the inferior margin (fig. 4 *A*). Furthermore, the third infuscated band transverses the wing at an angle of approximately 20 degrees or less in *completa*, while in *suavis* the angle has not been ob-

served to be less than 40 degrees, and is commonly less acute. The shape of the wings in these two species differs somewhat, particularly in that a slightly increased degree of tapering toward the apex produces more of a point in *suavis* than in *completa*. The claspers of *completa*, viewed meso-laterally, are somewhat curved and taper gradually from the center toward the distal end, while those of *suavis* are only very slightly curved and taper abruptly at the distal end. The ejaculatory apodeme of the internal male genitalia of these two insects presents important differences. In *completa* the distal portion of this structure is broadly rounded and flattened, while in *suavis* it is club-shaped (fig. 5) and only about one-third as wide at its greatest diameter as in the former.

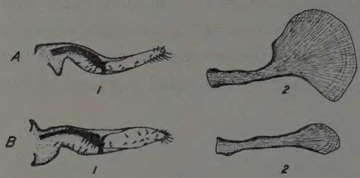



Fig. 5. Comparison of size and shape of claspers (1), and ejaculatory apodeme (2), of *Rhagoletis completa* (*A*), and *R. suavis* (*B*).

Fig. 6. *Rhagoletis completa*, female, dorsal aspect, showing yellowish-white scutellum, wing markings, characteristic position of wings, and extended ovipositor.

The female (fig. 6) differs from the male only in its slightly larger size, more pointed abdomen, and the presence of the ovipositor.

*Egg.*—The egg is somewhat curved in shape (fig. 7). It is pearly white when first deposited, becoming darker as the embryo develops. The posterior end tapers slightly and terminates in a very short pedicel, while the anterior end is more pointed. Fine reticulations occur over the entire surface of the shell, though they are more dense on the posterior end. The measurement of 100 eggs supplied the following data: average length 0.96 mm, range in length 0.8 to 1.16 mm; average width 0.22 mm, range in width 0.21 to 0.26 mm.

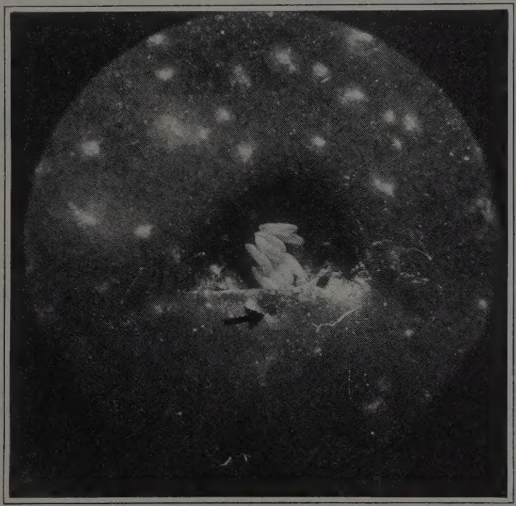


Fig. 7. Photomicrograph of *Rhagoletis completa* eggs in husk tissue of walnut. Arrow indicates puncture made by female in ovipositing.

*Larva.*—First-instar larvae that are ready to molt range from 1.8 mm to 2.0 mm in length and 0.4 mm to 0.6 mm in width. The body is nearly transparent (fig. 8 *A*), and the large cephalo-pharyngeal skeleton and oral hooks, or mandibular sclerites, are conspicuous because of their dark color (fig. 9 *A*). Each hook bears a prominent tooth-like process located on the blade about midway between the tip and base (fig. 9 *A*). The tracheal system is distinctly visible in detail (fig. 8 *A*). A longitudinal trunk begins at each posterior spiracle and extends forward to the prothoracic segment where it terminates. Anterior spiracles in this instar are lacking. Each posterior spiracle consists of two stigmatic plates, or peritremes through which air enters the trachea (fig. 10 *A*).

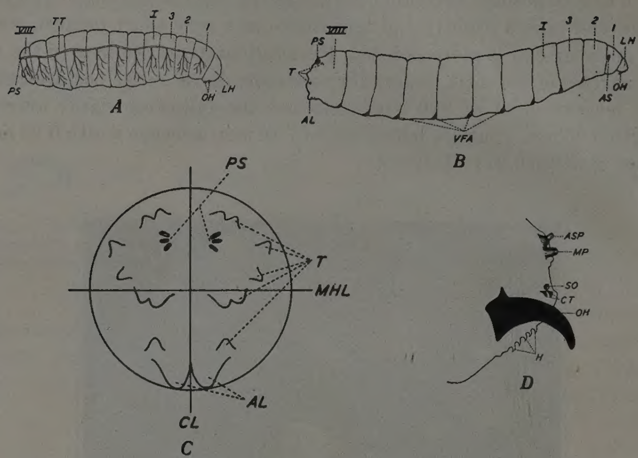



Fig. 8. Structural characters of *Rhagoletis completa* larvae: *A*, first instar, lateral view; *B*, third instar, lateral view; *C*, posterior end, third instar; *D*, head, third instar, lateral view. AL, Anal lobe; AS, anterior spiracle; *ASP*, anterior sense papilla; *CL*, center line; *CT*, chitinized teeth; H, hooklets; LH, larval head; MHL, mesohorizontal line; MP, maxillary palpus; OH, oral hooks; PS, posterior spiracle; SO, sense organ; T, tubercles; TT, longitudinal tracheal trunk; VFA, ventral fusiform areas; 1, prothorax; 2, mesothorax; 3, metathorax; *I*, first abdominal segment; *VIII*, eighth abdominal segment.

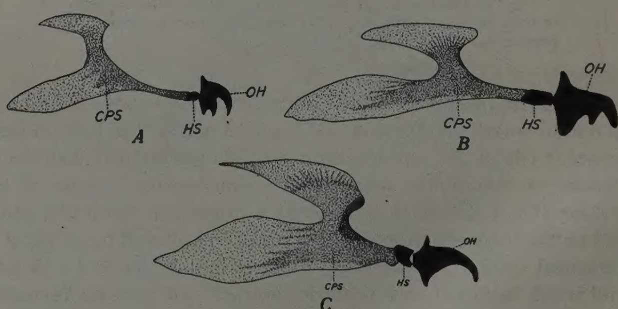



Fig. 9. Mouth parts of *Rhagoletis completa* larvae: *A*, first instar; *B*, second instar; *C*, third instar. CPS, Cephalo-pharyngeal skeleton; HS, hypostomal sclerite; OH, oral hook, or mandibular sclerite.

Four groups of spine-like projections which Efflatoun[13] refers to as "interspiracular processes" are evident on each spiracle.

Second-instar larvae that are about ready to molt range from 4.0 mm to 4.5 mm in length and 1.0 mm to 1.2 mm in width. The body is whitish in color and semiopaque, though dark contents of the alimentary canal are plainly observable. The tooth-like process on each oral hook is retained (fig. 9 *B*). Anterior spiracles occur on the posterior lateral portion of the prothoracic segment, one on each side. They are

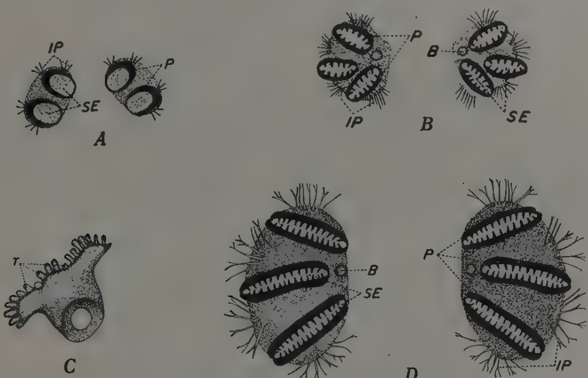



Fig. 10. Spiracles of *Rhagoletis completa* larvae: *A*, first instar, posterior; *B*, second instar, posterior; *C*, second instar, anterior; *D*, third instar, posterior. B, Button; IP, interspiracular processes; P, peritremes; SE, spiracular entrances; T, tubules.

yellowish, somewhat fan-shaped, and possess from 15 to 20 stigmatic papillae, or tubules, arranged in an irregular row on the distal margin (fig. 10). The presence of these spiracles readily distinguishes second-instar larvae from those of the first instar. Each posterior spiracle consists of three peritremes instead of two as previously indicated for first-instar larvae. These structures are yellowish, ovate, and the angle at which each is located with respect to a horizontal plane is characteristic of this species (fig. 10).

Mature third-instar larvae (fig. 8 *B*) average approximately 9.0 mm in length by 2.0 mm in width and they are yellowish-white in color. The tooth-like projection on the blade of each oral hook is absent (fig. 9 *C*). Several prominent structures, other than the oral hooks, occur on the head (fig. 8 *D*). The antennae, or anterior sense papillae, are short two-jointed organs situated on the anterior portion of the head, one on each side. A single-jointed structure, bearing several sensory rods near the

apex, is located just below each antenna. Snodgrass[36] considers these organs as posterior sense papillae, while Efflatoun[13] considers them as maxillary palpi. A pair of small chitinized teeth occur on the side of the mouth, immediately laterad of each oral hook. A sense organ is situated directly above each pair of these teeth, while below them, on each side of the mouth, is a short row of nonchitinized hooklets. The anterior spiracles differ from those found in second-instar larvae in size only. The posterior spiracles are orange-yellow and are slightly raised from the body surface. The peritremes differ somewhat in general shape and relative position to the horizontal plane from those of second-instar larvae (fig. 10 *B* and *D*). The angle at which the lower peritreme is situated with respect to the horizontal appears to characterize this species. The interspiracular processes situated outside and between each peritreme are evident as variable, branching spine-like projections. Ventral fusiform areas are present on nine consecutive segments beginning with the metathorax (fig. 8 *B*). The posterior body segment bears fourteen tubercles, which are characteristically situated and are of considerable importance taxonomically. These structures are shown in figure 8 *C*, following the diagrammatic method used by Greene.[19] Eight tubercles are located above the center line, and the dorsal ones are bifid. Six occur on the lower half of the segment, and those directly below the posterior spiracles are bifid.

*Pupa.*—The pupa is somewhat barrel-shaped, straw-colored, and measures approximately 5 mm in length by 3 mm in width (fig. 11). Because of telescoping of the anterior body segments of the larva when the puparium was formed, the anterior spiracles project conspicuously and are dark brown. Both anterior and posterior larval spiracles may be viewed from the dorsal surface. The lateral spiracles of the adult respiratory system are evident on the mesonotum and all abdominal segments. The circular cleavage line is located approximately in the middle of the first abdominal segment and encircles the entire puparium, though it is most prominent on the dorsal half. The horizontal cleavage line connects with the circular cleavage line at right angles on each side and extends around the anterior portion of the puparium in a midlateral plane. On the venter the larval mouth and anal opening appear as dark-brown, invaginated areas. The ventral fusiform areas are present, though not conspicuous.

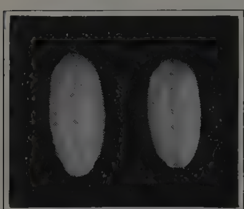


Fig. 11. Pupae, dorsal aspect (left), and ventral aspect (right).

## RELATED SPECIES ATTACKING WALNUTS

Four members of the genus *Rhagoletis* have as primary hosts species of *Juglans*. All are probably indigenous to the continent of North America. The insects are: *R. completa, R. suavis, R. juglandis,* and *R. boycei.* A brief discussion of the last three species follows:

*Rhagoletis Suavis.*—*Rhagoletis suavis* (Loew) (fig. 12) is the largest species of the genus and was described in 1862 by Loew[24] under the old genus *Trypeta.* It is commonly known as the walnut husk maggot.

Fig. 12. *Rhagoletis suavis,* male, showing characteristic wing markings and yellowish-white lateral stripe on otherwise yellow thorax.

Prior to 1930 this species was recorded from Massachusetts, Connecticut, New York, Pennsylvania, Maryland, West Virginia, North Carolina, South Carolina, Ohio, Indiana, Illinois, and Minnesota. Since then new records have been obtained, in which instances preserved specimens of larvae or adults, or both, have been determined by the author. These records, together with the authority, are as follows: Manhattan, Kansas, 1930 (R. Smith); Ames, Iowa, 1930 (Beck); Sturgis, Mississippi, 1930 (Myers); Bloomfield Hills, Michigan, 1931 (Ries); Fayetteville, Arkansas, 1931 (Baerg); and Columbia, Missouri, 1932 (Haseman). The present known distribution is shown in figure 13. The distribution of this insect probably conforms to the range of the eastern black walnut, *Juglans nigra,* and the butternut, *J. cinerea.* Brooks[8] has also

reared this insect from the Persian walnut, *J. regia,* and the Japanese walnut, *J. sieboldiana.* It is reported to be of economic importance in New York,[17] Pennsylvania, and Maryland in relatively small plantings of Persian walnuts. This species would probably constitute an economic problem should it become established in the commercial walnut-producing areas of the West.

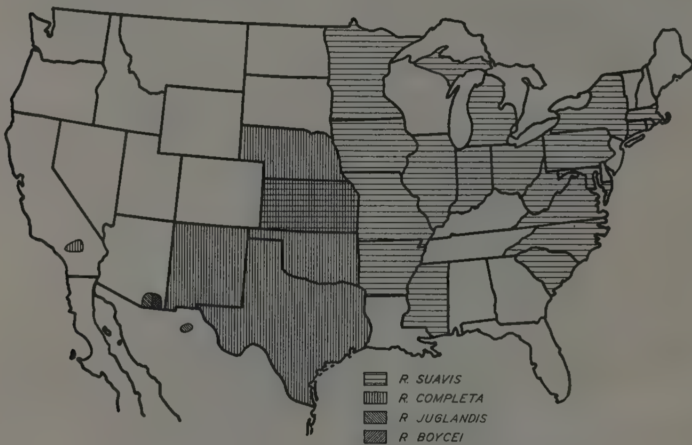



**Fig. 13. Distribution of the walnut-inhabiting *Rhagoletis.***

*Rhagoletis Juglandis.—Rhagoletis juglandis* Cresson (fig. 14) was described in 1920 by Cresson[11] from material collected by C. R. Biederman, the larvae of which were feeding in the green husks of Persian walnuts, and Arizona black walnuts, *Juglans rupestris,* in Carr Canyon, Huachuca Mountains, Arizona. Van Duzee collected this fly from Badger, Arizona. There are no other records from the United States, and those reported are close together and within a few miles of the Mexican border. Beck collected this insect on black walnut at Colonia Dublan, Chihuahua, Mexico, in 1931 (fig. 13). It is probably of Mexican origin. The adult is considerably smaller than *suavis* or *completa* and almost entirely yellowish in color. Observations on its field behavior, made in this study, indicate that this species is the most active one of the group attacking walnuts. When visiting Carr Canyon in July, 1930, J. C. Caldwell found that the adults had apparently emerged earlier than usual. He obtained second-instar larvae that were feeding in the developing walnut kernel since the shell had not hardened sufficiently to

prevent penetration. It appears that this species is probably capable of causing serious losses if established in commercial walnut-producing areas.

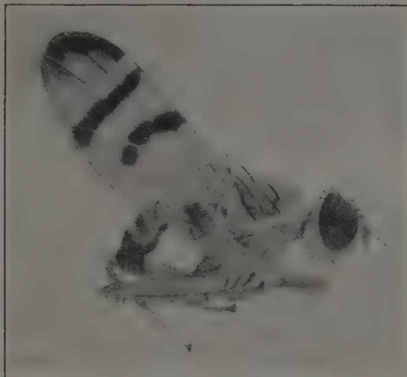


Fig. 14. *Rhagoletis juglandis,* male (topotype), showing relatively small size, characteristic wing markings, and yellowish-white lateral stripe on otherwise yellow thorax.

Fig. 15. *Rhagoletis boycei,* male (topotype), showing relatively large size, characteristic wing markings, and yellowish-white lateral stripe on otherwise black thorax.

*Rhagoletis Boycei.*—*Rhagoletis boycei* Cresson (fig. 15) was described by Cresson[12] in 1929 from material collected by K. L. Wolff and the author from Carr Canyon, Huachuca Mountains, Arizona (fig. 13).

Here it occurred with *juglandis,* though only a few specimens were observed at that time. In one instance a female was collected while ovipositing in the green husk of a Persian walnut, which fact indicates that this walnut is probably a host. Little is known regarding the distribution and biology of this insect. Limited observations in the type locality indicate that it is very wary and wild, and possibly has habits approaching solitude. Therefore an opinion regarding its possible economic importance is withheld.

## COMMON NAME

The matter of a satisfactory common name for *Rhagoletis completa* has received much consideration. Early in this study, when the species had been determined erroneously as *R. juglandis,* it was evident that the common name "black walnut fly" by which it had already been referred to in literature[11] was not suitable. A committee was appointed by the Entomological Club of Southern California to consider common names and was instructed to submit the most satisfactory name to the American Association of Economic Entomologists for adoption. In view of the generally accepted concepts of a satisfactory common name, and because a "walnut husk maggot" already existed, the name "walnut husk fly" was proposed and was officially accepted.[1]

In the light of present knowledge, this official common name was actually intended for *Rhagoletis completa,*[5] instead of *R. juglandis.* The author accepts this version of the matter, and the insect is known as the walnut husk fly among entomologists, walnut producers, and others who have occasion to use a common name in California.

## DISTRIBUTION

*Rhagoletis completa* is apparently indigenous to mid and south central United States (fig. 13) and its known distribution does not extend very far east of the one hundredth meridian. Since the correct identity has been established,[12] knowledge regarding distribution has been greatly increased. At the time of Cresson's study of this species from California in 1929, it was known, though erroneously determined, from but one other state—Texas. It was collected at Brownwood, Texas, by A. I. Fabis, in 1917, and from Pecan Bayou, Texas, in 1918 by the same collector. The coöperation of an entomologist in each state of the Union was solicited, and as a result new records were secured for this species as well as for *suavis.* In all instances the coöperators have kindly forwarded preserved specimens of larvae or adults, or both, to the author for determination. The records obtained for *completa,* and the authority, are as follows: Lincoln, Nebraska, 1930 (Swenk); Manhat-

tan, Kansas, 1930 (R. Smith); Stillwater, Oklahoma, 1931 (Sanborn); Comanche, Texas, 1930 (Nickels); and State College, New Mexico, 1932 (Eyer). Entomologists report personal knowledge of the existence of this species in Kansas, Oklahoma, and Texas for thirty to fifty years. *R. completa* has not been collected east of the ninety-fifth meridian, and there is but one record of *suavis* west of the ninety-seventh. At Manhattan, Kansas, both species were taken from the same tree. The ratio of *completa* to *suavis* adults obtained from extensive rearings there in 1932 was approximately 50:1. This probably indicates the operation of factors which limit the western distribution of *suavis*, while conditions for *completa* apparently approach optimum. In all records outside California the host of *completa* has been listed as wild (black) walnut; however several species are probably involved.

In California the insect is recorded from Los Angeles, San Bernardino, and Riverside counties. A relatively small area in each county is infested, the total constituting approximately 500 square miles. The total infested acreage of commercial walnut groves at the end of the 1932 season was approximately 2,000 acres. The locations of the first recorded infestation in 1926, together with other data regarding distribution, are shown in figure 1.

## WALNUT AS A HOST

"Host" as used in this study means any plant that serves to support the insect under any condition through the egg and larval stages, provided larvae so developed produce normal pupae and adults.

This species is practically monophagous, confining its attack almost entirely to the genus *Juglans*. During the course of these investigations infestations have been recorded from every species of *Juglans* and from practically all varieties of Persian walnut, *J. regia*, found growing within the infested area. The wild species and hybrids are: southern California black walnut, *J. californica;* northern California black walnut, *J. hindsii;* eastern black walnut, *J. nigra;* paradox hybrid walnut, *J. regia* × *J. hindsii,* and Royal hybrid walnut, *J. hindsii* × *J. nigra.* The cultivated varieties of Persian walnut are listed in two groups according to the degree of susceptibility to attack.

| Very susceptible | Slightly susceptible (resistant) |
| --- | --- |
| Eureka | Placentia |
| Franquette | Seedling (most types) |
| Mayette | Ehrhardt |
| Klondike | Ware |
| Payne | Neff |
| Seedling (certain types) | |

*Husk Hardness as a Factor in Varietal Susceptibility.*—Early in the history of the walnut husk fly in California, a very pronounced difference in varietal susceptibility to attack was observed. An instance was recorded in 1928 in which over 95 per cent of the walnuts on Eureka trees were infested, while those on Placentia trees alternately planted among the Eureka trees were only 2 per cent infested. Observations indicated that the later-ripening, thick-husked varieties were most favored as hosts. Since the flies oviposited in the green husk tissue, varietal susceptibility appeared to be related to the hardness of the husk at the time of oviposition activity. In order to obtain information regarding this matter, studies were conducted to determine the pressure in grams required to puncture the husks of the several varieties of walnuts during the oviposition period.

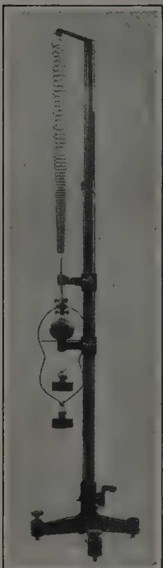


Fig. 16. Jolly balance modified for use in determining the pressure required to puncture green husks of walnuts and the skins of various fruits.

*Method of Testing Husk Hardness.*—A modified Jolly balance (fig. 16) was used to determine husk hardness. This instrument consists essentially of a spring with one end attached to the arm of an upright extensible column while the other end is attached to a steel puncturing rod which also has weights indirectly connected by means of a stirrup-shaped frame. The puncturing rod assembly has a horizontal hair line on that portion which connects to the lower end of the spring. This portion of the rod passes through a short piece of glass tubing that is mounted on the frame and which also has a hair line at its center. The puncturing rod used was cylindrical with a tip that was flat in cross section. A stage on the upright column supports the material that is to be tested for hardness. The extensible column is graduated and a vernier scale is mounted on the stationary column to permit accurate reading.

The technique of operation is as follows: Tension is applied on the spring by extending the inner portion of the upright column by means of a rack and pinion adjustment. When the tension is sufficient to balance the attached 400-gram weight plus the weight of puncturing rod and rigging, adjustment is continued until the hair line on the metal rod coincides with that on the glass tube through which it is suspended. The walnut to be tested is placed on the stage. Then the stage is adjusted so that the tip of the puncturing rod is barely touching the spot

on the husk to be punctured. With the walnut held firmly in place the tension on the spring is steadily reduced until the rod penetrates the husk. Penetration is indicated by a sudden drop of the flat-tipped rod into the tissue.

The reading on the graduated column at the point where penetration occurred is taken and subtracted from the zero (reading when spring tension perfectly balances weight, and hair lines on metal rod and glass tube coincide), which permits the calculation of the weight in grams required to puncture the green husk. When the cross-sectional area of the puncturing rod is known, the actual pressure in grams required per square millimeter to puncture the husk may be computed. The rods used in 1929 and 1930 became inadvertently mixed with others before their respective areas were computed. Therefore the data given for husk hardness during those two years are only relatively comparable to other data presented. However, the same rod was used throughout a single season, and thus all the data for any one season are strictly comparable. In the 1931 and 1932 studies the same rod was used throughout and the area computed, thereby permitting comparison of husk hardness on the basis of grams pressure required to puncture one square millimeter of husk surface. The area of the puncturing rod was 0.2243 sq. mm.

In the tests of 1929 and 1930, samples of 25 walnuts were selected at random from each variety. Two puncture readings were made of each walnut, one for the stem region and one for the middle region. Thus 50 readings were made per test for each variety. Preliminary puncture data showed that the calyx region was materially harder than other regions; therefore this region was disregarded in these tests. In the tests of 1931 and 1932 each random sample of a variety consisted of 50 walnuts, and 12 punctures were made per walnut, 4 punctures placed in each of the stem, middle, and calyx regions. The punctures in each region were equally spaced on the circumference. Thus the number of readings per test per variety was 600.

*Results of Husk-Hardness Tests in 1929.*—The data obtained in 1929 are graphically presented in figures 17 and 18.

Figure 17 shows that the husks of Placentia walnuts are considerably harder than those of the Eureka variety. The former is one of the least susceptible, while the latter is one of the most susceptible varieties. The reason for the softened condition of the husk during the middle of August is not known. Husks soften as the walnuts approach the ripening condition. Most of the eggs were deposited during late August and the fore part of September (fig. 59). Normal harvest for the Eureka variety begins about October 15, while the harvest for the Placentia variety begins about September 15. Therefore the period intervening

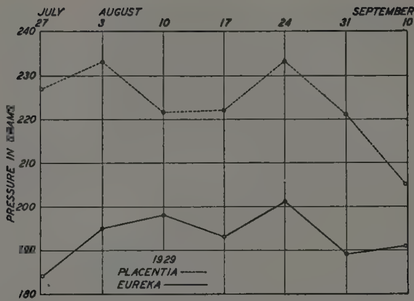



Fig. 17. Pressure in grams required to puncture the green husk of Placentia and Eureka walnuts during the period of activity of *Rhagoletis completa* (1929).

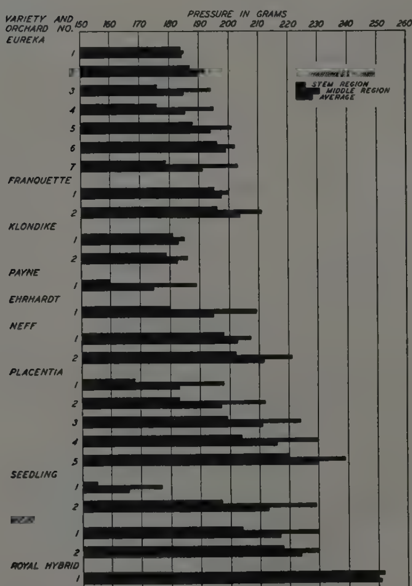



Fig. 18. Husk hardness of various walnut varieties, together with variation in hardness of the same variety in different groves. Data collected August 25–28, 1929.

between the time that Placentia husks reach a susceptible state and the time of harvest is too short for appreciable larval development.

The data presented in figure 18 show a wide range of husk hardness with different groves of the same variety. The differences are probably due to differences in orchard management together with local conditions influencing the developing crop. From figures 18 and 59 it is evident that oviposition in Eureka walnuts took place when a pressure of approximately 190 grams or less was required to puncture the husk. On this basis the walnuts in several orchards of susceptible varieties were not in a susceptible condition at this time and subsequent observations showed them lightly infested. Previous observations had shown that occasional orchards of susceptible varieties were lightly infested despite the presence of many flies. Furthermore infestation of the Franquette variety usually takes place later than that of other varieties. The data indicate that certain resistant varieties, such as Ehrhardt, Placentia, and Seedling, were in susceptible condition for infestation. In these instances the early harvest would have prevented any appreciable amount of larval development.

It is particularly interesting to note that in every instance with the Persian walnut the stem region was softer than the middle region. Oviposition data in 1928 showed that 70 per cent of the egg cavities were located in the stem region.

*Irrigation and Susceptibility to Infestation.*—Field observations and limited field data indicate that, other factors being comparable, irrigation practices have a bearing on susceptibility to infestation. An abundant supply of water throughout the growing season apparently increases susceptibility. In 1927 and 1928 the trees nearest irrigation outlets and those at the low end of the run of water where flooding takes place, were the first infested and were usually more heavily infested than those receiving less water. Furthermore several instances are recorded where nuts on trees of the Placentia variety (normally resistant) were apparently rendered susceptible to the extent that 25 per cent or more were infested as a result of being located within a few feet of a leaky irrigation stand. The inference was that perhaps abnormal amounts of water resulted in more succulency in the tree, rendering the husk softer and thereby creating a condition more favorable to oviposition. In one instance data obtained from trees in a certain grove of the Eureka variety where the soil received excessive amounts of water at each irrigation showed that an average pressure of 187.6 grams was necessary to puncture the husks while 203.1 grams was necessary where trees received normal amounts of water. Further studies regarding irrigation and susceptibility to attack were conducted in 1931.

*Husk-Thickness Studies in 1929.*—Limited studies were made in 1929 on husk thickness of the more common varieties. A steel millimeter scale with beveled tip was employed for this purpose. The tip of the scale was inserted into the green husk, at a right angle to the surface, until the shell of the nut was reached, at which point the thickness of the husk was read in tenths of a millimeter. The measurements were made on the same walnuts that were used in the husk-hardness tests. Two measurements were taken in each stem and middle region. Preliminary measurements showed that the husk was consistenly thinnest in the calyx region. Data were not collected for this region since eggs are rarely deposited in this location. A mean of 100 readings per variety constituted the data presented in figure 19.

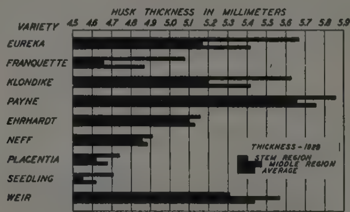



Fig. 19. Husk thickness of the common walnut varieties in the area infested by *Rhagoletis completa.* Data collected August 25–28, 1929.

The data do not conclusively show that a relation exists between husk thickness and susceptibility to infestation. However, it is interesting to note that the most resistant varieties possess a thinner husk than the very susceptible ones, and furthermore that the ratio of thickness of stem region to middle region in the very susceptible varieties is generally greater than in the resistant ones. The fact that approximately 70 per cent of the egg cavities observed were located in the stem region indicates the possibility of a relation between thickness and location of egg cavity which would suggest the general relation between thickness and hardness.

*Results of Husk-Hardness Tests in 1930.*—Data were obtained in 1930 regarding the husk hardness of the most important commercial varieties in both infested and uninfested walnut-producing areas of the state. These data are presented in figure 20. In all instances the data show that the stem region is softer than the middle region.

Conclusions regarding the probable degree of susceptibility of varieties growing in different sections of the state are unwarranted. The time

of ripening of any given variety changes with the locality. Therefore important differences in husk hardness at the time of oviposition are to be expected. Furthermore, the effect of climatic conditions in various sections of the state upon the time of emergence of the fly is unknown. Should flies emerge in early June it is probable that the walnut husks would not have attained sufficient hardness to prevent oviposition. More-

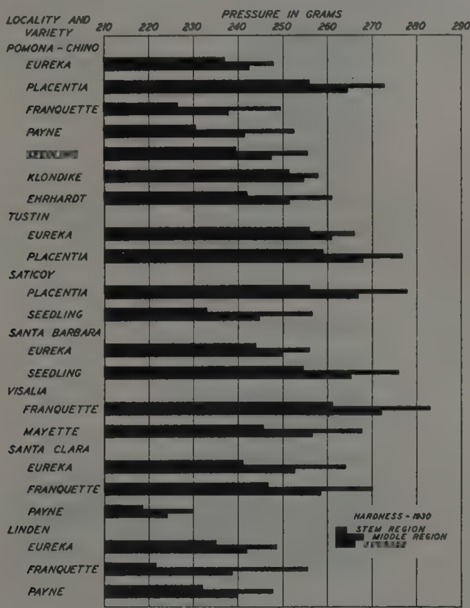



Fig. 20. Husk hardness of the more important varieties of Persian walnuts from the walnut-producing areas of California. Data collected August 20–25, 1930.

over if the time of adult emergence was not materially altered it is entirely probable that the most susceptible varieties growing in those sections where maturity is reached relatively later in the season may be attacked before the husk hardens sufficiently to prevent oviposition. Should the walnut become infested before the shell is formed and hardened, the larvae would no doubt consume the kernel in feeding.

*Results of Husk-Hardness Tests in 1931.*—The husk-hardness studies were continued in 1931 and were considerably enlarged upon with respect to the effect of irrigation practices upon husk hardness and subsequent degree of susceptibility of Eureka walnuts to infestation. These

husk-hardness irrigation data more particularly concern economic control and are for that reason treated elsewhere under 1931 field control plots, experiment XVII, page 549. The data of 1931, comparing husk hardness of various varieties, are presented in figure 21.

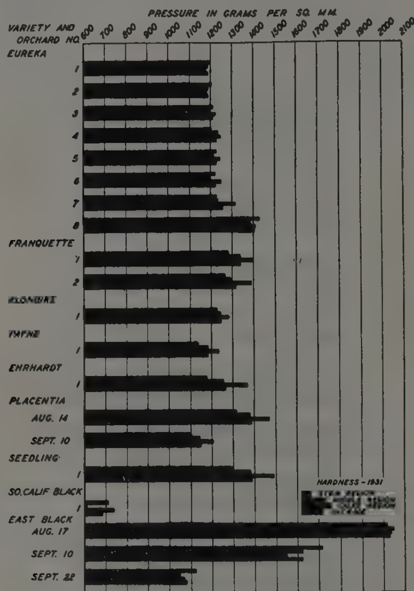



Fig. 21. Husk hardness of various varieties of walnuts, showing variation in hardness in various groves and on different dates. Data collected August 18–22, 1931, unless otherwise indicated.

The Eureka walnuts from various orchards were remarkably uniform in husk hardness with the exception of Orchard No. 8. This orchard adjoins Orchard No. 1 on the south. It is of especial interest to note that irrigation is not practiced in Orchard No. 8, while it is an important factor in the orchard practices of most commercially produced walnuts. The degree of infestation in Orchard No. 8 was very light, despite the fact that large numbers of flies were liberated in the grove experimentally. Observations in this grove over a period of five years have shown that a consistently light infestation exists even when the infestation in the adjoining grove has on several occasions reached 90 per cent.

The data given for the Placentia variety are from the same grove, though obtained on different dates. They show that the husk becomes appreciably softer as the nuts approach ripening. A similar relation is shown by the data for the eastern black walnut.

In most instances the 1931 data for the Eureka variety indicate very slight differences in hardness between the three regions of the husk. Furthermore, in most instances the middle or calyx region is softer than the stem region, which is contrary to the 1929 and 1930 data. With the other Persian varieties the ascending order of hardness is stem, middle, and calyx regions, which is in accord with data obtained in previous years.

*Results of Husk-Hardness Tests in 1932.*—The observed differences in hardness of the various regions of Eureka walnut husks led to further studies in 1932. A comparison was made between the hardness of the three husk regions of the Placentia and Eureka varieties extending from July 16 to September 7. The data obtained, indicating the trend of husk hardness throughout the seasonal activity of the fly, are presented in figure 22.

The data for 1932 show that the stem region in Eureka walnuts is appreciably harder than either the middle or calyx regions, while the reverse condition exists with the Placentia variety. The average hardness of the latter variety is materially greater than that of the Eureka variety. This fact substantiates the earlier work of 1929 and is probably the most important factor governing the wide difference in susceptibility to attack of these two varieties.

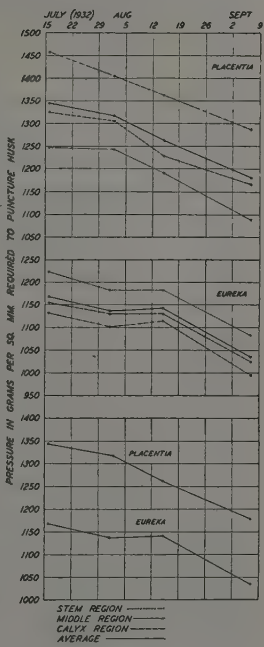



Fig. 22. Husk hardness of Placentia and Eureka walnuts in 1932.

*Importance of Husk Hardness as a Factor in Susceptibility.*—The most important factor pertaining to the susceptibility of varieties of Persian walnuts to attack by the walnut husk fly is that of hardness of the husk. Husks of Persian walnuts increase in hardness as the nuts increase in size and age, reaching the peak of hardness usually in late June, after which they become softer as maturity is approached. The degree of hardness when the peak is reached and the extent of subsequent softening appears to be a varietal

characteristic. As a result of the physical nature of the husk of the resistant varieties, the fly is unable to penetrate the surface in order to deposit eggs. Under both laboratory and field conditions females have frequently been observed to fail in their attempts to puncture the husks of the less susceptible varieties. Furthermore they are unable to puncture the husks of susceptible varieties until a certain degree of maturity and resultant softness has been reached.

Owing to individual variation in nuts upon a single tree with respect to husk hardness, some nuts are in a susceptible condition throughout the entire period of fly activity. The female fly apparently finds the susceptible walnuts and also the location for the egg cavity by "trial and error." Females are frequently observed attempting oviposition at many places on walnuts without success, before finally succeeding in penetrating the husk of a walnut and depositing a batch of eggs.

## PEACH AS A HOST

From laboratory rearing records and certain field studies the peach, *Amygdalus persica,* has proved to be a host. Eggs were deposited and larvae reached maturity in the Elberta, Simms, Lovell, and Phillips varieties in a battery-jar cage. Also the immature stages were completed when adults were confined on a tree of the Lovell variety by means of a $12 \times 12 \times 12$ foot cheesecloth cage over the entire tree. The varieties mentioned mature relatively late in the season during the height of fly activity.

Flies were commonly observed on peach trees in the vicinity of infested walnut trees. Natural infestation has been recorded in several instances where peaches were growing as interplants with Eureka walnut trees that were infested. However, the average number of larvae maturing in individual peaches under both field and laboratory conditions was relatively few, which indicates that this host does not afford optimum conditions for development. Adults emerging from larvae that developed in peach were apparently normal in all respects. It appears that at least some peaches from trees growing interplanted with infested walnuts are harvested with eggs and young larvae in them. The period elapsing between the time when peaches are in a physical condition favorable to oviposition and the time of normal harvest is too short for an appreciable amount of larval development to take place. The observed infestations under natural conditions were in tree-ripe peaches that had not been harvested. Hardness tests on unripe peaches from one interplanted grove showed that 245 grams pressure was required for penetration, while 193 grams was required for the Eureka walnut inter-

plants. At that time the peaches were not infested but the walnuts were. Later, as the peaches approached ripeness and became softer, they were infested, but no data on hardness were obtained.

## STUDIES ON POSSIBLE HOSTS

Laboratory studies were conducted with the object of determining whether or not certain fruits would serve as hosts for *Rhagoletis completa.* In a few instances tests were conducted in the field. The general procedure of the laboratory tests was to confine from 25 to 50 gravid females and the same number of males in an inverted battery-jar cage containing the fruit to be tested. Usually 12 tests were made with each fruit. Sucrose was included in each cage as food. Each test extended over a period of several weeks. The behavior of the flies in the various tests was observed several times daily. The host material was removed periodically and inspected for the presence of eggs. When oviposition had occurred the fruit was placed in a battery jar and kept under conditions favorable for the development of the insect. Sifted sand was placed in the bottom of each battery jar so that any larva maturing could pupate. The sand was again carefully sifted at the termination of the test to note whether or not pupae were present.

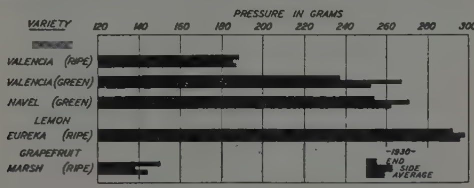



Fig. 23. Rind hardness of certain citrus fruits. Data collected September 17, 1930.

Several fruits and vegetables commonly grown in the infested district were tested.

*Orange.*—The navel orange (*Citrus sinensis*) is eliminated as a possible host because it ripens in the winter season and the green oranges are too hard for the females to puncture, as shown by data on skin hardness (fig. 23) and by laboratory studies of the behavior of adults when confined with green navel oranges.

In numerous laboratory experiments conducted with ripe Valencia oranges, sexually mature flies were confined for weeks with the oranges. In every instance females oviposited, though at no time were the eggs inserted deep enough in the skin to hatch before they dried up. They

punctured the rind of the orange rather readily (fig. 24) and went through the actions of normal oviposition, except for the fact that they were unable to lacerate the inner tissue of the rind to make a cavity in which to place the eggs. Instances were observed where females worked vigorously for half an hour trying to make an egg cavity, without success. Often a single egg was deposited in a puncture and others were placed on the surface near the hole.

In several of the tests Eureka walnuts were placed in the cage with the oranges. The flies remained on the oranges almost exclusively,

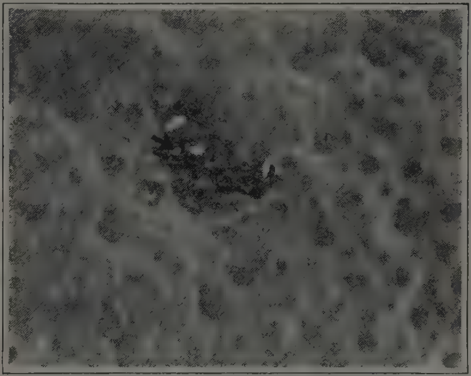


Fig. 24. The surface of a Valencia orange, with punctures representing many attempts to oviposit; and four eggs, three deposited on surface and one, indicated by arrow, nearly submerged in rind tissue.

actively attempting oviposition and directing no attention to the walnuts until the oranges were removed from the cages.

In a field test a cheesecloth cage ($12 \times 12 \times 12$ feet) was built over a small Valencia orange tree bearing a good crop of ripe and also green fruit. A total of 500 flies, some newly emerged and some sexually mature, was liberated on this tree at varying intervals of time. To insure an adequate supply of food, sucrose-sweetened water was sprayed on the foliage. The flies appeared to behave normally. At the end of the season all oranges were sliced and examined for the presence of larvae. None was found; however, a few oranges exhibited evidence of attempts at oviposition by the females.

In laboratory tests involving a total of 50 oranges, eggs were artificially placed in the tissue below the skin. In 4 of these hatching took place and the larvae reached maturity. In 1 orange several larvae pu-

pated within the tissue, while the others emerged through the hole where the eggs were injected. When early-stage larvae were placed within orange tissue they apparently developed in a normal manner and reached maturity. The foregoing data indicate that the Valencia orange might serve as a host if females were capable of depositing their eggs normally. However, it should be pointed out that the identity of pupae of larvae

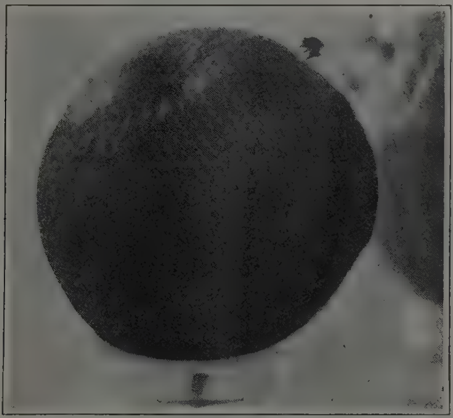


Fig. 25. Tangerine, showing female ovipositing in ripe fruit.

that developed in orange tissue was inadvertently lost, so that it is not definitely known whether or not adults emerged from this particular material.

The Mediterranean Sweet variety was tested in a limited manner. When the oranges were dissected one contained small larvae. Closer examination showed that a female had successfully placed her eggs in the orange to a depth adequate for hatching.

*Tangerine.*—Females were successful in ovipositing (fig. 25) in this loose-skin type of citrus fruit (*Citrus deliciosa*). The eggs were deposited beneath the skin, though not into the fleshy pulp tissue. While hatching occurred, larvae were not found in any of the fruits examined, and none reached maturity in these tests. Since eggs or small larvae were not artificially placed in the flesh tissue, it is not known whether or not larvae are capable of developing in this medium.

*Grapefruit.*—Both Marsh and Duncan grapefruit (*Citrus grandis*) were placed in cages with ovipositing females. The actions of the flies were similar to those reported for the Valencia orange. One fruit in par-

ticular exhibited eleven "brown spot" areas when removed, indicating many attempts at oviposition (fig. 26). Hardness data (fig. 23) showed that the skin of grapefruit was considerably softer than other species of citrus tested; however, no eggs were deposited deep enough to insure hatching.

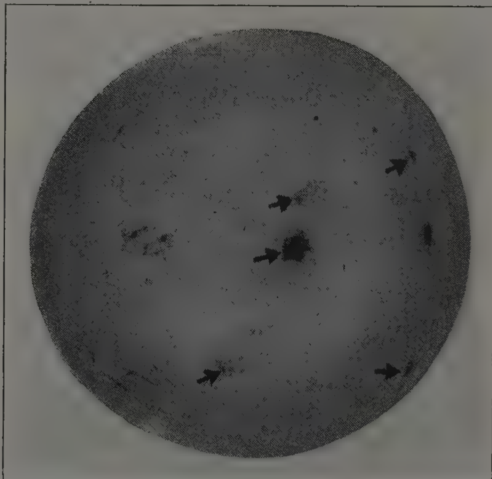


Fig. 26. Grapefruit, showing points where many unsuccessful attempts at oviposition were made.

*Lemon.*—Eureka and Lisbon lemons (*Citrus limonia*) were both tested under laboratory conditions. Females repeatedly attempted oviposition without success. In twelve lemons of each variety, eggs were artificially inserted. The final check of these failed to reveal that any larvae had matured; in fact none was found, though indications of larval feeding were noted in several fruits.

*Apple, Pear, and Quince.*—The Jonathan and White Pearmain varieties of apple (*Pyrus malus*), the Bartlett pear (*Pyrus communis*), and the Orange quince (*Cydonia oblonga*) were used in laboratory tests. Eggs were deposited in all these species of pome fruits and hatching occurred; however, in no instance did larvae reach maturity. Flies have been commonly observed on these fruit trees where they were growing interplanted with infested Eureka walnut trees; however, neither eggs nor larvae have been found in any of the fruits under such conditions.

*Plum.*—Most of the plums (*Prunus salicina*) growing in the infested area are harvested before the peak of seasonal activity of the fly. How-

ever, a late-maturing strain of the Satsuma variety was tested. Of the twelve plums placed in cages with flies, eggs were deposited in five. These eggs hatched, but none of the larvae reached maturity.

*Fig.*—The Kadota fig (*Ficus carica*) was tested rather extensively. Many branches bearing figs in various stages of development were placed in cages with adults. The females attempted oviposition in many instances; however, no eggs were deposited. The milky, viscous exudation emanating whenever the skin is punctured possibly is repulsive to the fly. Females have been observed to withdraw their ovipositor almost immediately after the puncture is made and they apparently have considerable difficulty in cleaning it.

A cheesecloth cage (12 × 12 ×12 feet) was built over a small tree heavily laden with fruit, and a total of 500 flies was introduced at varying intervals. At the end of the season, no larvae or pupae were found.

*Grape.*—The Tokay, Muscat, and Emperor grapes (*Vitis vinifera*) were tested in the laboratory. In one instance a female was observed to attempt oviposition but no eggs were found in any of the grapes, and at the end of the experiment, no larvae or pupae were found.

*Prickly Pear.*—Females oviposited readily in the cactus fruits (*Opuntia* sp.). In these tests it was very apparent that the flies did not care to rest on the fruit since they were usually noted there only when ovipositing. On dissecting several fruits it was noted that the eggs had hatched, but none of the larvae reached maturity.

*Pecan.*—Green fruits of both seedling and budded pecans (*Hicoria olivaeformis*) were tested. Flies frequently attempted oviposition, but in no instance were they successful in penetrating the husk to deposit eggs.

*Sierra Sweet Bay.*—Sierra sweet bay (*Myrica hartwegii*) occurs commonly in the canyons of the Sierra Madre and has many of the attributes of the Juglandaceae—in fact the families are closely related. A limited number of the nutlets were placed in a cage with flies very late in the season. Oviposition was not observed, and neither larvae nor pupae were found at the end of the experiment.

*Avocado.*—Several Mexican varieties of avocados (*Persea gratissima*) that ripen in the early fall and a later variety, the Fuerte, were placed in cages with flies. Females were observed to attempt oviposition without success; the fruits, which were green, possessed a hard skin. Neither larvae nor pupae were found at the end of the experiment.

*Pomegranate.*—Ripe pomegranates (*Punica granatum*) were placed in cages with flies. Several attempts at oviposition were observed, though in no case were the females able to insert their ovipositors into the skin.

*Potato.*—The Idaho Russet potato (*Solanum tuberosum*) was used in these tests. In three instances eggs were deposited beneath the surface of the tuber, and a few eggs were placed on the surface near the punctures. Examination six weeks later showed that the eggs had hatched but only one larva was found. This particular larva was in the second

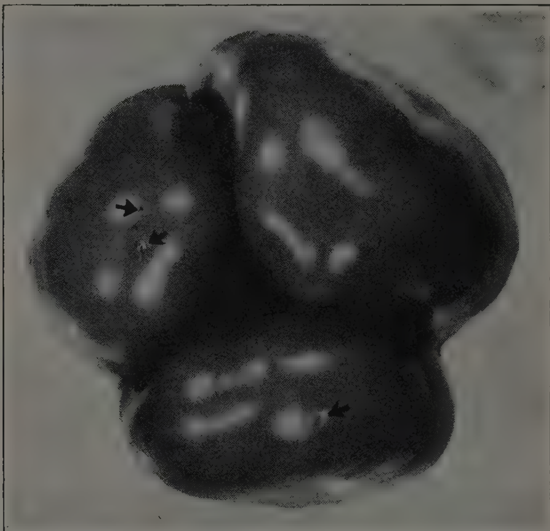


Fig. 27. Bell pepper: the arrow at the upper left indicates cavity of eggs deposited normally; other arrows indicate eggs on surface above cavities that are filled with eggs.

instar and died before maturing. This test was conducted in an effort to find a host which could be used for continuous rearing of the insect in the laboratory.

*Eggplant.*—Many instances were recorded of females ovipositing in the mature eggplant (*Solanum melongena*). One female died with the ovipositor inserted. The eggs hatched but none of the larvae reached maturity.

*Bell Pepper.*—The flies oviposited very freely in mature bell peppers (*Capsicum grossum*). There were several cavities of eggs in each pepper tested (fig. 27). In one instance a batch of nine eggs was found adhering to the surface of the pepper near a cavity containing eggs. None of the larvae reached maturity.

*Tomato.*—Ten ripe and twenty green tomatoes (*Lycopersicum esculentum*) were used in these tests. Eggs were deposited in all tomatoes used and in some cases several batches were found in each. When eggs were deposited in green tomatoes, a callus formed around and below the puncture in the tissue. The eggs hatched but most of the larvae died before maturing. However, two larvae reached maturity and formed normal-sized pupae. When adults failed to emerge the pupal cases were opened and the insects found to be dead. It is questionable whether or not mortality was due to environmental conditions after pupation or to the effect of the host during larval development.

*Discussion of Studies on Possible Hosts.*—While most of the host studies were preliminary in nature, they are indicative of the fact that the flies will attempt to deposit their eggs in many kinds of fruits under artificial conditions. In some instances degeneration of the fruit undoubtedly affected the development of the young larvae. Thus it appears that, given natural host conditions, some larvae would probably mature in certain of the fruits tested.

The insect has never been observed even attempting to oviposit in the stem of a plant, whether succulent or otherwise. Therefore, the larvae appear to be entirely restricted to inhabitation of fruits.

During the five years that the insect has been studied, most species of fruits and vegetables growing within the infested area have been rather carefully observed for indications of infestation. None has been noted to be infested, with the exception of walnut and peach.

The extent to which fruits other than the preferred host, walnut, will be attacked under natural conditions is problematical. However, the observations and experimental data to date indicate that the species is not likely to become of economic importance on other crops.

## INJURY AND ECONOMIC IMPORTANCE

The degree of infestation is dependent upon a combination of factors and may vary from less than 1 per cent to over 95 per cent of the nuts on the tree.

*Shell Stains.*—Injury caused by *Rhagoletis completa* is manifested in several ways. The principal type is shell stain, resulting from the feeding of the larvae within the exocarp or green husk of the nut. The surface of the husk directly above the feeding tunnels assumes a black color caused by the decay that takes place beneath (fig. 28). This blackened area increases as the larvae extend their feeding range and in many cases includes the entire husk, though usually about one-half or less of it is affected. The juices from the soft decay of the inner husk permanently

darken the shell of the nut (fig. 29). This staining is probably caused by tannin released from the broken-down tissue, and in many instances it extends entirely through the shell.

In laboratory experiments, normal newly harvested walnuts were partially submerged in tannic acid solutions of varying concentrations and lengths of exposure. Characteristic staining of the submerged por-

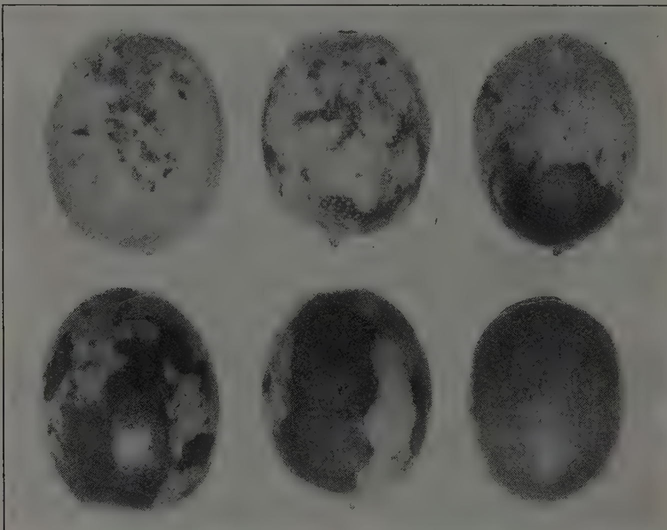


Fig. 28. Characteristic external appearance of injury by larvae feeding within husk. Note that injured husk tissue has not shrunk, and that a definite line marks the color changes from black to normal green.

tions of the shells was produced with concentrations as low as 1 per cent when exposed for 48 hours. In an attempt to remove the stain from infested walnuts, experiments involving the use of chloroform, ether, sulfuric acid, chloride of lime, potassium permanganate, chlorine, and ethylene were conducted with negative results. Tests with sand blast were also carried out without success. Any nut having a shell stain which is not removed by the regular chlorine bleaching process in packing-house operations becomes a "cull."

Since there is no known method of removing the stain resulting from infestation by this insect, nuts so infested are classified as culls, and the returns to the producer are appreciably reduced as a result. Such cull walnuts are normally processed in a cracking plant and the carefully

graded meats marketed in bulk under trade names representing their quality. Packing-house records show that cull walnuts net the producer approximately 50 per cent less than normal walnuts, even though they contain unimpaired kernels. The Eureka variety, which is very susceptible to infestation, usually commands the highest market price.

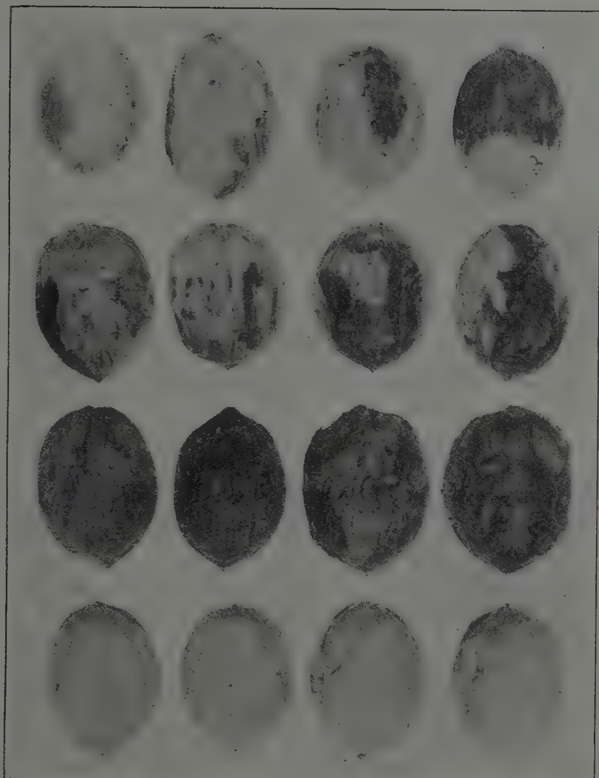


Fig. 29. Upper three rows show varying amounts of shell surface stained as a result of infestation. Lower row shows uninfested nuts.

All infested walnuts, however, do not become culls, and when they are not classed as culls it is doubtful whether or not an economic loss results. Usually from 10 to 25 per cent of the nuts attacked exhibit no evidence of having been infested after they are hulled at harvest. The extent of injury is somewhat dependent upon seasonal conditions with

reference to the development of the host and the emergence and subsequent development of the immature stages of the fly.

*Reduction in Quality of Kernels.*—The secondary type of injury includes a reduction in quality of some of the kernels. According to the

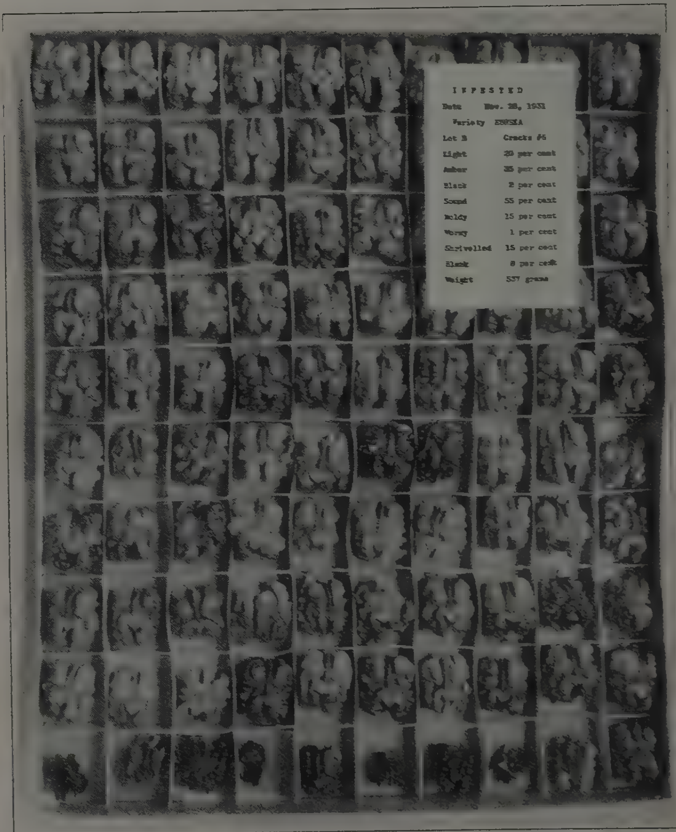



Fig. 30. Typical lot of 100 infested walnuts showing standard method of grading kernels in order to determine the extent of injury resulting from infestation.

"grades" established by the California Walnut Growers' Association, the value of walnuts is primarily dependent upon a clean, light-appearing shell, and such factors pertaining to the kernel as insect damage,

TABLE 1

EXTENT OF INJURY TO WALNUT MEATS DUE TO INFESTATION BY RHAGOLETIS COMPLETA, 1929–1931, AS SHOWN BY CRACK TESTS

| Year and group* | Light | Amber | Black | Moldy | Shrivel and blanks | Sound | Weight | Merchantable† | Grade after culling† | Returns to producer† (orchard run) |
|---|---|---|---|---|---|---|---|---|---|---|
| | *1* | *2* | *3* | *4* | *5* | *6* | *7* | *8* | *9* | *10* |
| | *per cent* | *per cent* | *per cent* | *per cent* | *per cent* | *per cent*‡ | *grams* | *per cent* | | *cents per pound* |
| 1929 | | | | | | | | | | |
| *A* Normal | 85 | 6 | 0 | 0 | 9 | 91 | 565 | 94 | Diamond | 18.9 |
| *B* Infested | 72 | 13 | 1 | 5 | 9 | 85 | 555 | 86 | Diamond | 17.8 |
| *B* compared with *A* | −13 | +7 | +1 | +5 | 0 | −6 | −10 | −8 | .......... | −1.1 |
| 1930 | | | | | | | | | | |
| *A* Normal | 38 | 28 | 1 | 0 | 33 | 66 | 459 | 73 | Emerald | 13.1 |
| *B* Infested | 40 | 25 | 1 | 1 | 33 | 65 | 455 | 74 | Emerald | 13.3 |
| *B* compared with *A* | +2 | −3 | 0 | +1 | 0 | −1 | −4 | +1 | .......... | +0.2 |
| 1931 | | | | | | | | | | |
| *A* Normal | 44 | 38 | 1 | 2 | 15 | 82 | 540 | 89 | Emerald | 13.6 |
| *B* Infested | 16 | 42 | 6 | 13 | 23 | 58 | 522 | 64 | Suntand | 9.3 |
| *B* compared with *A* | −28 | +4 | +5 | +11 | +8 | −24 | −18 | −25 | .......... | −4.3 |
| Average, 3-year period | | | | | | | | | | |
| *A* Normal | 56 | 24 | 1 | 1 | 19 | 80 | 521 | 85 | .......... | 15.2 |
| *B* Infested | 43 | 27 | 3 | 6 | 22 | 69 | 511 | 75 | .......... | 13.5 |
| *B* compared with *A* | −13 | +3 | +2 | +5 | +3 | −11 | −10 | −10 | .......... | −1.7 |

* Each group consisted of 10 lots of 100 walnuts each.

† Data calculated by A. W. Christie, Field Manager, California Walnut Growers' Association, from data supplied in other portions of this table, together with available packing-house records.

‡ Columns 1+2+3+4+5=100; columns 6+3+4+5=100.

color, shriveled condition, and presence of mold. Larvae feeding within the green husk apparently affect the kernel with respect to certain of these factors, particularly in those nuts attacked earlier in the season. Since the juice that causes the stain usually penetrates the entire thickness of the shell it probably darkens the meat in some instances. Because the largest percentage of eggs is deposited in the stem region of the husk, the larvae first attack the tissue in that region. Consequently some of the conductive tissue transporting nutritive substances to the developing kernel is destroyed. It seems probable that under certain extreme conditions shriveling of the kernel results. Humidity conditions conducive to the growth of fungi on the kernels probably exist as a result of infestation, particularly when larvae have tunneled the entire husk and the ensuing damp, mushy decay is present. Thus when accompanied by protracted climatic conditions favorable to fungi development, mold is likely to become evident on the kernels.

Crack tests were made to ascertain the effect of infestation on the quality of the kernels (fig. 30). In order to obtain a significant comparison, random samples of 1,000 nuts exhibiting evidence of having been infested, and another of noninfested nuts, were taken from the same grove at harvest. These samples were dehydrated and otherwise handled under identical conditions. The results of these crack tests are presented in table 1.

When the various factors affecting quality of the walnuts in normal and infested lots are averaged for the three-year period, it is evident from the data in table 1 that color of kernel, moldiness, proportion of blanks, and weight are appreciably affected by infestation. The data show an average reduction in merchantable kernels of 10.5 per cent, with the resultant reduction of 11.5 per cent in net returns to the producer. The loss ranges from 0 to 25 per cent. The data given with respect to percentage of merchantable nuts and cents per pound net to producer (orchard run) were calculated from the crack-test records, and the injury resulting from stained shells in the infested lots was ignored. By treating the data in this manner, it is possible to arrive at the reduction in value of the walnuts due to infestation, aside from the primary type of injury. Therefore, by closely estimating and calculating, the total reduction in net returns to the producer from injured nuts amounts to from 50 to 75 per cent, allowing 50 per cent for the primary type of injury and from 0 to 25 per cent for the secondary type.

*Harvesting Costs.*—Harvesting costs are increased somewhat by the presence of infested walnuts. The husks of the majority of the infested nuts do not split normally and allow the nut to drop (fig. 31). Thus more time is consumed at harvest in shaking the tree to remove these

"sticktight" walnuts. After these infested walnuts are on the ground, the adhering husks must be removed by hand labor or by hulling machinery. Furthermore when freshly hulled, infested walnuts are put into sacks or piles with uninfested nuts, some staining of the shells of the latter occurs.

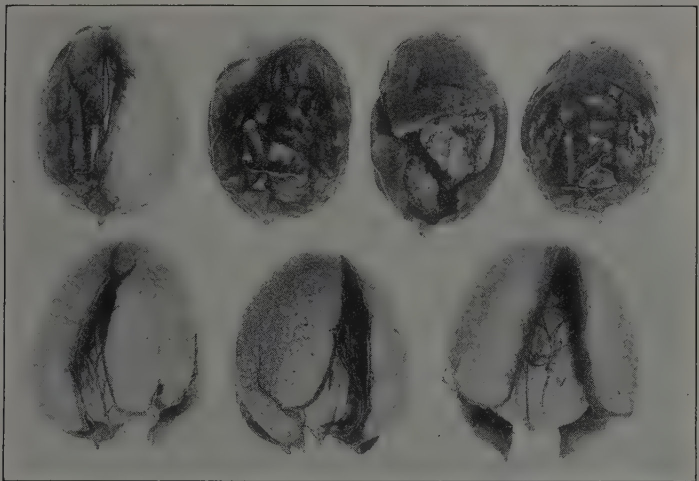


Fig. 31. Upper row, "sticktights" produced by infestation. Lower row, uninfested walnuts, showing how husk normally splits.

*Incidental Expenses.*—Other economic considerations are the costs incident to enforcement of quarantines for the prevention of artificial spread. This is particularly important since approximately 30 per cent (30,000 acres) of the total walnut acreage in California is planted to susceptible varieties, most of which is not within the present infested area.

## LIFE HISTORY AND HABITS

The treatment of life history and habits in this study deals with each stage of the insect in the sequence of natural occurrence. To conserve space many of the detailed data are presented graphically only. Therefore the points in graphs were merely connected and no attempt was made to smooth curves. In many instances several sets of data are plotted on the same chart, thus precluding the inclusion of the smoothed curve together with that connecting the points.

## ADULT

*Cages Used in Studying Adult.*—During the early part of this study considerable difficulty was experienced in keeping adults alive for appreciable lengths of time in any type of container or cage other than inverted jelly glasses. Most of the workers on other species of *Rhagoletis* have reported similar experiences. The inverted jelly-glass type

Fig. 32. Inverted battery-jar cage and typical set-up used in biological studies. The section of petri dish contains absorbent cotton, saturated with liquid food. Jar extends beyond the edge of the plate-glass shelf, to permit aeration. The vent thus provided is covered with wire screen.

of cage was decidedly too small to carry on the desired studies. The success of such a cage appeared to be due to the maintenance of a humidity condition bordering on the optimum. It seemed evident therefore, that any type of cage that would duplicate the conditions of the inverted jelly glass would be satisfactory. Preliminary tests demonstrated the adaptability of inverted battery jars for these studies.

A description of the procedure employed throughout most of these studies follows: Battery jars 9 inches in height and 7¾ inches in diameter, with ground edge, were inverted on sections of plate glass ¼ inch thick, 9 inches wide, and 5 feet long (fig. 32). Sections of this length

were easily handled and kept clean. It was necessary to mount the plate glass on pedestals in crocks of water, thereby isolating the section and avoiding invasion by ants. One half of a 2½ inch petri dish, containing a wad of absorbent cotton thoroughly saturated with a 25 per cent sucrose solution, was placed in the cage to supply food and moisture. The inverted jar was left near enough to the edge of the plate glass to permit an opening of approximately ½ inch at the widest point.

The flies to occupy the cage were drawn into the glass tube of a regular suction-type catcher and then discharged into the inverted jar. This type of insect catcher was used exclusively for such handling of flies to minimize injury. When the desired number of flies had been injected

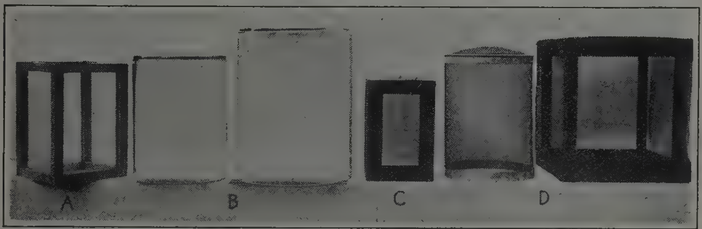



Fig. 33. Types of cages used in biological studies: *A*, quartz glass; *B*, flint-glass battery jars; *C*, 16-mesh wire-screen cage for suspending in foliage of trees; *D*, 16-mesh wire-screen cages used in laboratory.

into the cage, a small piece of 16-mesh screen was placed under the edge of the jar to prevent their escape. The weight of the jar held the screen in place. The vent prevented condensation of moisture on the sides of the jar during the day. Droplets of moisture were usually present on the walls of the jar during the night, but these in no way interfered since the flies did not move about to any appreciable extent after dark. In all cases, except in experiments where it was impracticable, such as the toxicity series, the jars and plate glass were washed once a week. This proved to be important in keeping flies apparently normal over periods of weeks. A jar of the size indicated will accommodate about 200 flies without undue crowding. It is believed that this type of cage was fairly satisfactory for laboratory handling of the flies with the possible exception of the effect of certain rays of the sun's spectrum. While there is practically no information available pertaining to this matter with regard to insects, it seems probable that beneficial light rays would be excluded by the type of glass from which the battery jars are manufactured. Limited studies were made with quartz-glass cages.

Other types of small cages were tried but none was satisfactory (fig. 33). Cages one foot square, completely covered with 16-mesh wire screen,

and cages with one half covered with wire screen and the remainder with cheesecloth, were tested extensively. In some instances improvised humidifiers, consisting of wicks or absorbent cotton in bottles of water, were used. The life of the flies was prolonged when the humidity was artificially increased; but still they did not behave normally. In only a few instances out of some 50 cages set up in this manner was oviposition obtained.

Of the larger types of cages over small trees the cheesecloth-covered cage was satisfactory, while the one covered with 16-mesh wire screen was not. Two walnut trees of the same variety were covered with 12 × 12 × 12 foot wire-screen cages, while two more trees were covered with cheesecloth cages of the same size. These cages were stocked with flies and otherwise managed under as nearly comparable conditions as possible. Flies remained alive over the entire season in the cheesecloth cages and infested 100 per cent of the nuts, while in the screen cages the flies were all dead at the end of four weeks, and most of them at the end of three weeks. The screen cages were restocked after four weeks. The degree of infestation of nuts was less than 50 per cent at the end of the season. It is interesting to speculate on the explanation for the difference in environment apparently created by the two types of cages. The humidity factor readily suggests itself, in view of the experience with smaller cages.

*Emergence from the Soil.*—In freeing itself from the pupal case in the soil to emerge, the adult pushes off the anterior end of the case. The point of rupture of the puparium is at the circular cleavage line in the middle of the first abdominal segment. Occasionally there is a rupture along the horizontal cleavage line also. The ptilinum no doubt plays an important role in forcing this pupal cap off. When the fly leaves the puparium the entire body is of a light color, very plastic, and capable of being greatly distorted. This faculty, together with the aid of the ptilinum, enables the fly to work its way through small cracks and other small places. As an example in many instances dead bodies of newly emerged flies have been found in the center of tightly rolled cotton plugs in glass vials in which pupae were being temporarily held. After the surface of the soil has been gained, the fly very diligently cleans itself. The ptilinum is inflated and deflated in the cleaning process, while being vigorously stroked with the forelegs which also serve to clean the antennae and mouth parts. On emergence the wings are folded along the longitudinal veins. The fly walks briskly about but stops frequently to stroke the much-folded wings and the abdomen with the hind legs. The three pairs of legs are cleaned by rubbing them together. In a total of over 50 flies observed emerging at different times, an average period of

20 to 30 minutes was necessary for the wings to become extended and hardened sufficiently for flight. The natural color pigments of the insect become conspicuous after several hours' exposure to daylight.

In connection with control experiments, detailed information regarding emergence was necessary. Therefore, the emergence studies were fairly extensive and extended from 1928 to 1932 inclusive.

*Methods Used in Emergence Studies.*—The cages used in these studies were 7 feet long, 6 feet wide, and 3 feet high, and were covered with


Fig. 34. *A*, standard type of adult soil-emergence cage ($7 \times 6 \times 3$ feet) in typical location under walnut tree. *B*, shelter containing thermograph for recording soil temperature within emergence cage.

cheesecloth (fig. 34). They were placed under infested trees, centered on the tree trunk, and extended parallel with the irrigation furrows. An effort was made to locate them at points representative of the particular area with reference to soil type, size of trees, and other factors. Since large numbers of flies were needed for other studies, the population of certain cages was augmented artificially in the following manner: A frame 6 feet long and 5 feet wide, with ¾-mesh chicken wire bottom, and mounted on legs 1 foot high, was placed over the site of an emergence cage. Infested walnuts were put into this frame and the larvae came out of the husks to enter the soil normally.

The cages were erected and covered with 50-mesh cheesecloth in June each year. This was early enough to catch the first emerging flies. An effort was made to ascertain the extent to which the cheesecloth cover-

ing altered the natural environment with respect to temperature and humidity. A hygrothermograph in an approved shelter was set up in a cage about one foot above the surface of the soil. This instrument had been carefully calibrated with the one to be set up under normal grove conditions. Furthermore, both instruments were checked by sling psychrometer readings at frequent intervals. Records from the emergence cage were kept for a period of three weeks. The comparison of conditions for a typical week is shown in figure 35.

The data show that in the emergence cages the temperature averaged approximately 1 degree higher at the upper limits and approximately

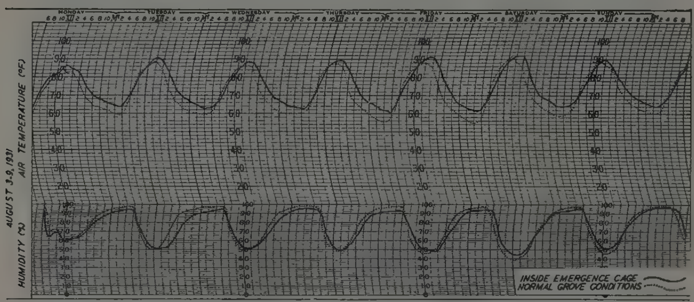



Fig. 35. Temperature and relative-humidity conditions existing inside an emergence cage in comparison with normal orchard conditions. (Typical 7-day period.)

2 degrees higher at the lower limits. The relative humidity varied inversely with the temperature to the extent of approximately 1 and 2 per cent, respectively. It seems improbable that the indicated differences in temperature and humidity between cheesecloth emergence cages and normal grove conditions influenced the emergence of adults.

Beginning with the emergence of the first fly of the season, each cage was visited daily at approximately the same time. The flies were carefully collected and a daily record was kept of total number of flies per cage with the ratio of females to males. Many different types of containers for collecting were tested in an effort to find the most satisfactory one from the viewpoint of speed in collecting, minimum injury, and ease of segregating and counting. A test tube, 15 mm in diameter, was found to be best adapted for use. Since the flies were usually on the walls and ceiling of the cage, the tube could be placed over a single fly very readily after a little practice. The insect usually landed in the bottom of the tube after the effort made to escape when the tube was placed over

it. However, the flies generally climbed upward on the walls as soon as equilibrium was gained, which made it necessary to jar the tube often by giving it a sharp rap against the palm of the hand. Fifteen to twenty flies were usually collected in one tube, which was then plugged with cotton and placed in the collecting box. When the box with tubes and flies was kept in the shade, the flies usually quieted down and by the time all were collected from the cage the ones taken earlier could be readily segregated and counted. The females were readily singled out because of the somewhat pointed abdomen.

The data pertaining to emergence each season are graphically presented and discussed, while a summary of the total emergence data concludes the treatment of this phase of the study.

*Definition of Terms Used in Emergence Studies.*—In discussing emergence several of the terms used require definition in order that the author's conception may be clearly understood. "Second generation" refers to those individuals that may emerge from the soil during the season in which they pupate. "Annual generation" refers to those individuals that remain in the soil throughout one winter, adults emerging the year after the season when larvae entered the soil and pupated. "Biennial generation" refers to those individuals that emerge the second year after pupation. "Multi-annual generation" refers to those individuals that remain in the soil longer than one year. "Seasonal peak" of emergence refers to the time when the greatest numbers of flies emerged. "Median" of emergence refers to the time when 50 per cent of the total number of flies have emerged.

*Emergence in 1928.*—The emergence data for 1928 were limited, since only four cages were employed. These were located in a grove that was moderately infested in 1927. There was considerable variation in the rate of emergence in the different cages. However, the variation among the four cages, two seeded and two natural, was not greater than the variation within either one of these groups. A total of 1,459 flies emerged; the sex ratio was 53 ♀ to 48 ♂. A composite of the data obtained is presented in figure 36.

Emergence began on July 12 and gradually increased until the seasonal peak was reached on August 18. Then a gradual decrease took place until the end of the season. The last flies were collected in the cages on September 25. Thus flies were emerging daily during a period of 75 days. The curve representing seasonal emergence may be considered a normal frequency polygon, which is the expected type, considering the nature of the phenomenon. The daily emergence record shows that females emerged in greater numbers than did males during the

fore part of the season, while the reverse was the case during the latter portion. Complete temperature records were not available during this season.

A cage of carefully sifted soil was maintained, which contained approximately 4,000 pupae of the earliest-maturing 1928 larvae. No flies emerged in this cage, indicating the absence of even a partial second generation.

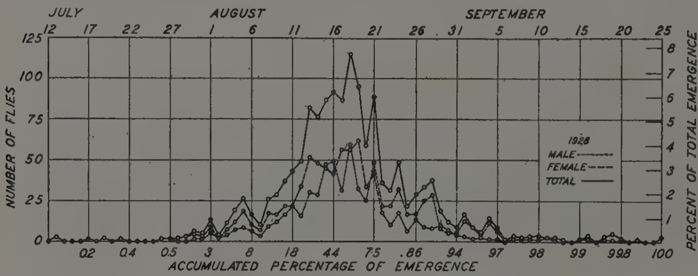


Fig. 36. Emergence of *Rhagoletis completa* in 1928.

*Emergence in 1929.*—A total of 20 cages supplied emergence data for the season of 1929. These cages were distributed over the infested area and were located in groves where a considerable degree of infestation existed in 1928. Hygrothermograph records furnished complete data regarding air temperature and humidity throughout the season. A total of 7,398 flies emerged. The sex ratio was 53 ♀ to 47 ♂. The general emergence data are shown in a composite chart, together with air temperature and humidity records, in figure 37.

Emergence began on July 19, reached the seasonal peak on August 26, and terminated October 9. Thus the total period of emergence was 82 days. The general shape of the emergence curve is similar to that of 1928. Likewise as in 1928, females were predominant in numbers during the fore part of the season, with the situation becoming reversed during the latter portion.

The mean daily air temperature and relative humidity shown in figure 37 represent an arithmetical mean of the readings at 2-hour intervals. Relative humidity apparently has no bearing upon emergence and is included on this chart only for reference convenience in dealing with other phases of the study. However, there is an indication of a relation existing between air temperature and emergence. Soil temperature is directly related to air temperature.

From the beginning of the emergence period until the seasonal peak is reached, the mean air temperature based on 5-day averages forms a

fairly smooth curve which corresponds to the curve of emergence of the flies. Soil temperature does not fluctuate so greatly nor so rapidly as air temperature; therefore it is unlikely that the several short periods of

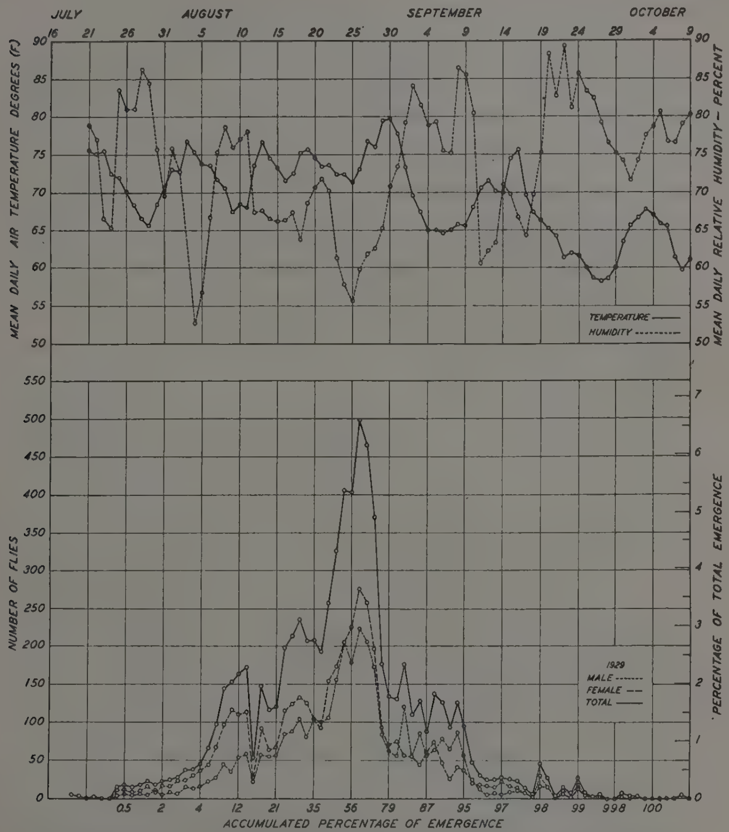



Fig. 37. Emergence of *Rhagoletis completa* in 1929, with air temperature and relative humidity.

lower air temperature just before the seasonal peak of emergence materially altered the temperature at a depth of several inches.

Data regarding time and rate of emergence of annual-generation flies in comparison with the biennial generation were obtained and are presented in figure 38.

A greater percentage of biennial-generation flies than of the annual-generation ones emerged early in the season. The accumulated emergence on August 23 was 94 per cent biennial in contrast to 43 per cent annual.

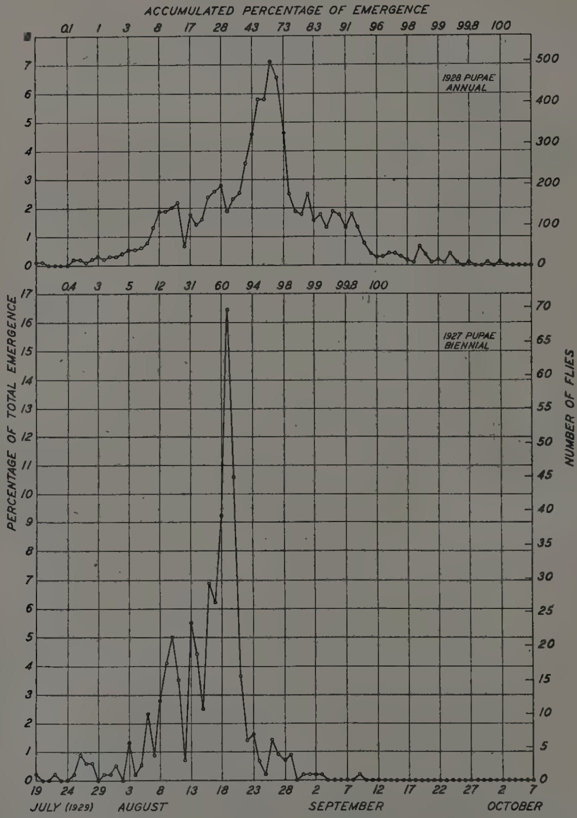



Fig. 38. Comparison of emergence of annual and biennial generations of *Rhagoletis completa* in 1929.

*Emergence in 1930.*—The emergence data for 1930 were obtained from 15 cages distributed in the infested area. A total of 8,965 flies emerged. The sex raio was 48 ♀ to 52 ♂. Throughout the period of emergence, thermographic records of soil temperature and hygrothermographic records of air temperature and humidity were maintained. The

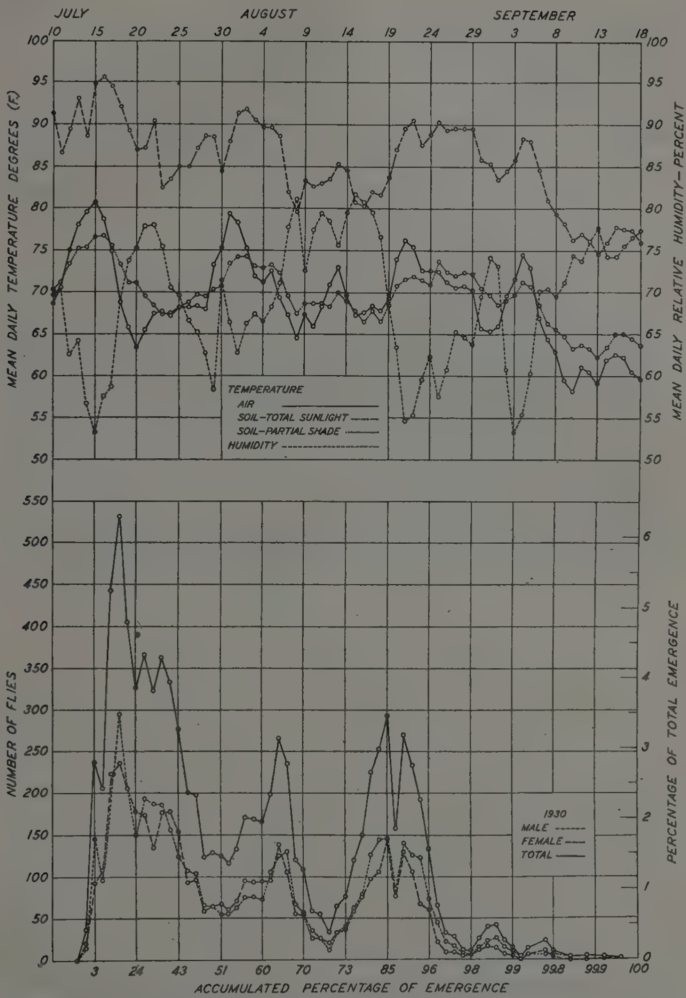



Fig. 39. Emergence of *Rhagoletis completa* in 1930, with prevailing soil temperature, air temperature, and relative humidity.

thermographs used were carefully calibrated at the beginning and were frequently checked throughout the season. The temperature of the soil at a depth of 3 inches in cages in total sunlight was compared with that of the soil in cages in partial shade at the same depth.

The data regarding fly emergence and records on air and soil temperature and relative humidity are presented in figure 39.

Emergence began on July 13, reached an early seasonal peak on July 18, and terminated on September 16. The total period was 65 days.

The records show only slight differences in the relative abundance of males and females throughout the emergence period. The general shape of the emergence curve differs greatly from those of the previous two years in that it is distinctly multimodal. This probably indicates the operation of certain factors affecting emergence heretofore not encoun-

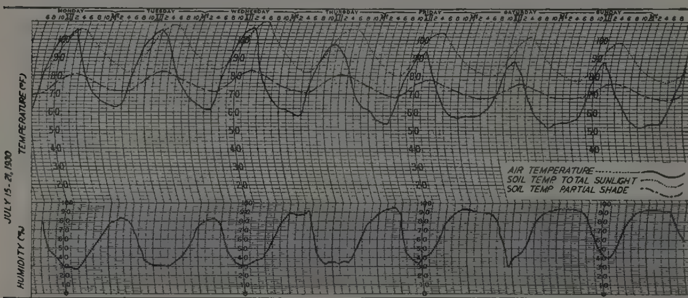



Fig. 40. Comparison of air temperature and soil temperature in cages in total sunlight, with soil temperature in cages in partial shade. (Typical 7-day period.)

tered, in which connection temperature is logically suspected. The soil-temperature data show a direct relation to emergence, since each major peak is closely followed by a major peak of fly emergence. The temperature of the soil in cages in total sunlight fluctuates only about one-half as much as air temperature. Soil temperature in cages in partial shade fluctuates slightly less than that in total sunlight. A lag of 2 or 3 days exists between important fluctuations in air temperature and the responding soil temperature. The records from charts of the three instruments for a typical week were transferred unaltered to one chart for comparative purposes. These data are presented in figure 40.

Data obtained regarding time and rate of emergence of annual-generation flies in comparison with the biennial generation are presented in figure 41.

Emergence of annual-generation flies began and reached the seasonal peak a few days earlier than in the biennial generation. However, after the first ten days of emergence the numbers issuing in both instances were fairly uniform. Minor seasonal peaks generally correspond in both cases.

Two cages containing approximately 3,000 pupae each, of the early-maturing 1930 larvae, were used to determine whether or not a partial second generation would develop. No flies emerged in either of the cages.

*Emergence in 1931.*—Twelve emergence cages were operated in 1931. A total of 11,856 flies emerged. The sex ratio was 47 ♀ to 53 ♂. The data obtained are presented in a composite chart in figure 42.

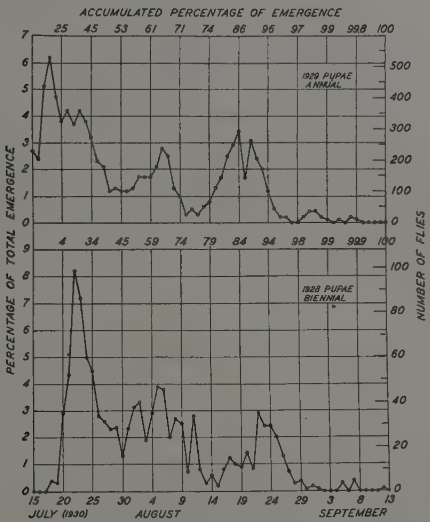



Fig. 41. Comparison of emergence of annual and biennial generations of *Rhagoletis completa* in 1930.

Emergence began on July 7, reached a very early seasonal peak on July 10, and terminated September 15. The total period of emergence was 70 days. Females and males emerged in about equal numbers throughout the emergence period. The general shape of the emergence curve is similar to that for 1930; therefore it likewise differs greatly from that for 1928 and 1929. The multimodal effect is apparently the result of the operation of factors similar to those that produced this condition in 1930.

Emergence began more abruptly in 1931 than in any preceding season. The mean daily temperature for the 10-day period prior to the beginning of emergence ranged from 75 to 79 degrees. Since these particular temperature data are not shown on the chart, the relation of temperature to the seasonal peak of emergence is not discernible. Sec-

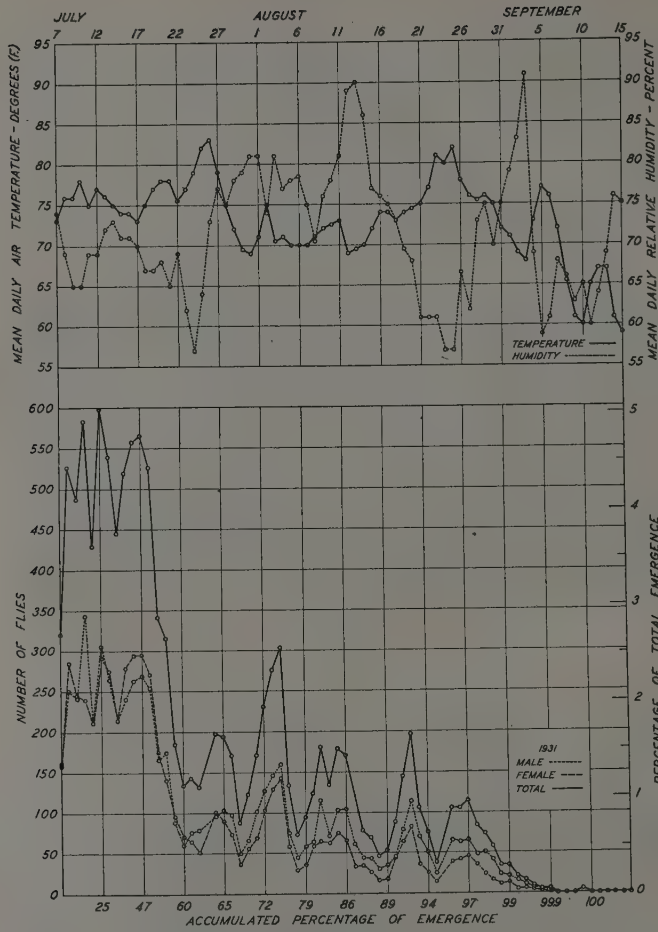



Fig. 42. Emergence of *Rhagoletis completa* in 1931, with air temperature and humidity.

ondary peaks of emergence rather closely follow rising temperatures above 70° F, indicating the relation of temperature to emergence.

Data obtained regarding time and rate of emergence of annual-generation flies in comparison with biennial-generation flies are presented in figure 43. A much greater percentage of biennial-generation flies

than of the annual-generation ones emerged early. In the former group the accumulated emergence on August 17 was 82 per cent, while in the latter it was 34 per cent.

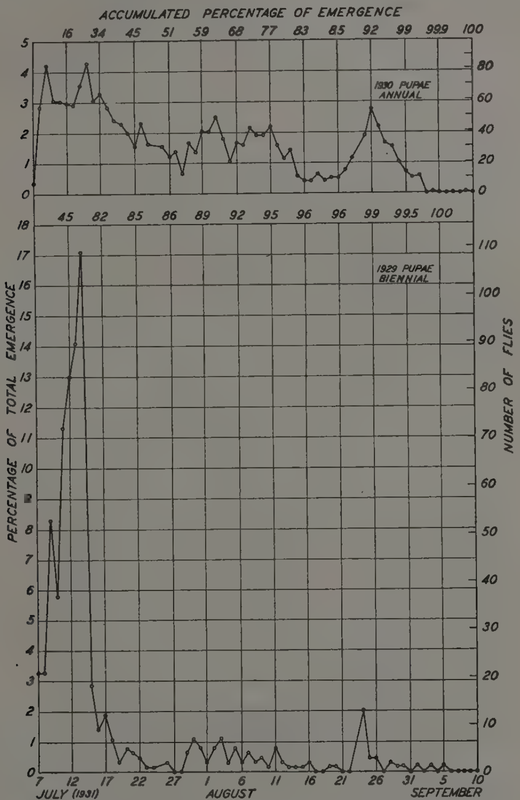



Fig. 43. Comparison of emergence of annual and biennial generations of *Rhagoletis completa* in 1931.

The time and rate of emergence from cages located on the north, south, and west exposures of trees were compared in 1931. These data are presented in figure 44. A material difference exists in the emergence of flies in the variously located cages. The emergence was most rapid on the west exposure, followed by the south and north. These differences are probably related to temperature.

*Emergence in 1932.*—Fifteen emergence cages were used in 1932. A total of 7,400 flies emerged. The sex ratio was 50 ♀ to 50 ♂. The data obtained are presented in figure 45.

Emergence began on June 29, gradually increased until August 10 when the seasonal peak was reached, and terminated on September 24.

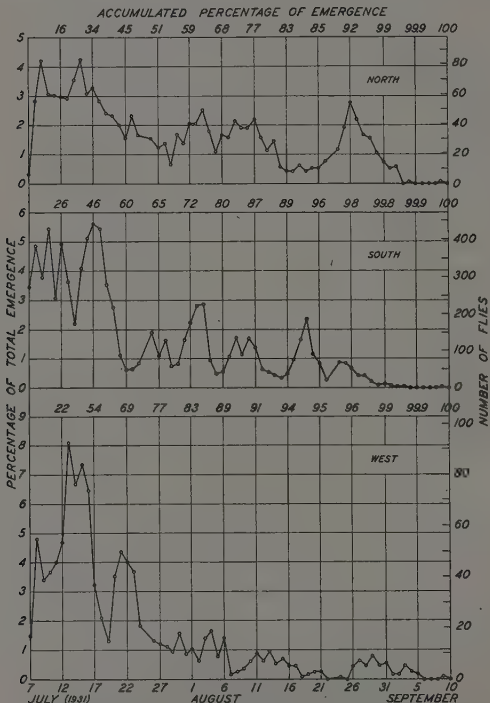



Fig. 44. Comparison of emergence of *Rhagoletis completa* in cages located on north, south, and west exposures of trees in 1931.

The total period of emergence was 96 days. Females were more abundant than males during the fore part of the season, while the situation was reversed during the latter portion.

The general shape of the emergence curve for this season more nearly approaches the normal frequency type than do those of 1930 and 1931 and in this respect is similar to that for 1928 and 1929. The relation of temperature to emergence is evident. The mean temperature averaged by 5-day periods during the fore part of the season increased gradually

and was accompanied by increased emergence. Just before the seasonal peak of emergence was reached, the temperature dropped; however, the soil temperature apparently remained sufficiently high to cause emergence to continue at a high rate for several days. The rapid drop in temperature probably produced the bimodal effect shown by the emergence curve.

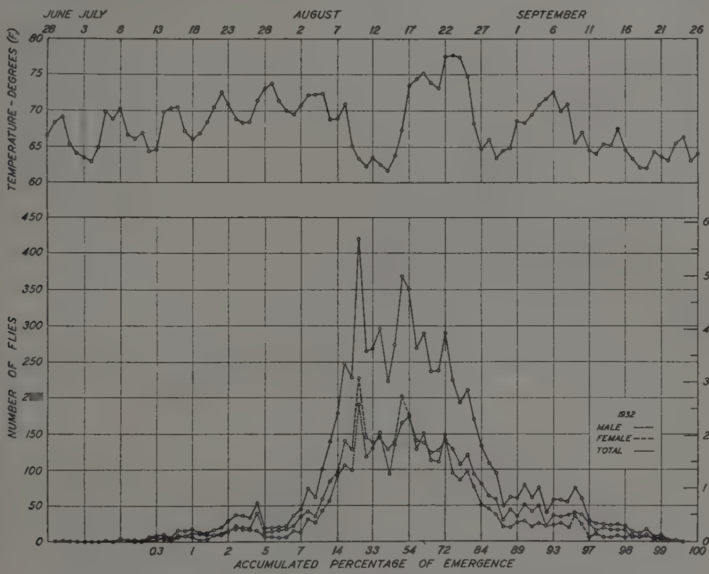



Fig. 45. Emergence of *Rhagoletis completa* in 1932, with air temperature.

Data obtained regarding time and rate of emergence of annual-generation flies and biennial-generation flies are presented in figure 46. A considerably greater percentage of biennial-generation flies than of annual-generation ones emerged early in the season. In the former group the accumulated emergence on August 7 was 71 per cent, in contrast to 25 per cent in the latter group.

Data regarding emergence from cages located on the north and south exposures of trees were obtained in 1932 and are presented in figure 47. Emergence began earlier in the season and the rate was more rapid in the southern cages than in the northern ones. The accumulated emergence on August 12 in the former was 37 per cent, while in the latter it was 13 per cent. The indicated differences are in accord with the 1931 data.

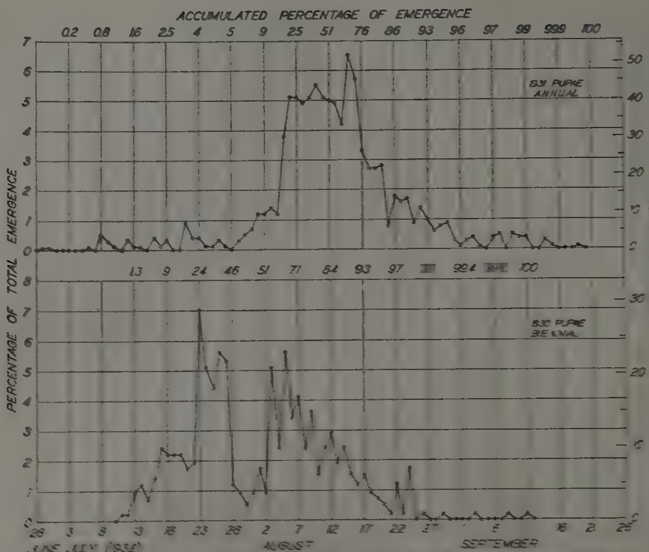



Fig. 46. Comparison of emergence of annual and biennial generations of *Rhagoletis completa* in 1932.

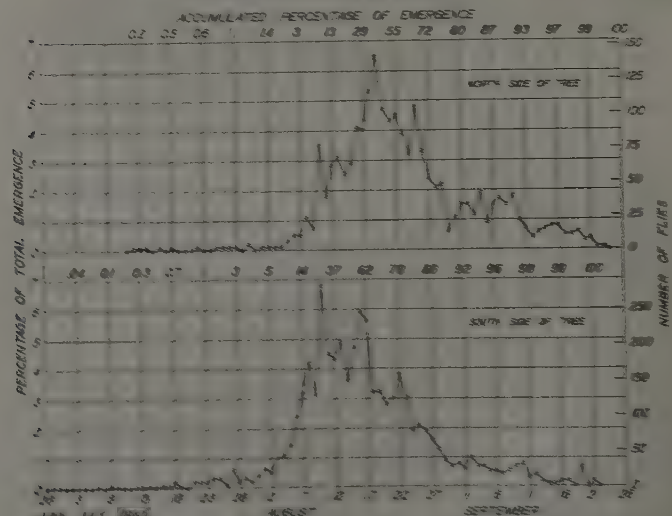



Fig. 47. Comparison of emergence of *Rhagoletis completa* in cages located on the north and south exposures of trees in 1932.

The effect of depth of pupae in the soil upon time and rate of emergence was obtained during 1932. In 1931, pupae were buried at a 3-inch depth in certain cages and at a 12-inch depth in others, in soil that had been freed of pupae by sifting. Emergence records obtained from these cages are presented in figure 48. The time and rate of emergence were materially increased in the cage 3 inches in depth over the cage 12 inches

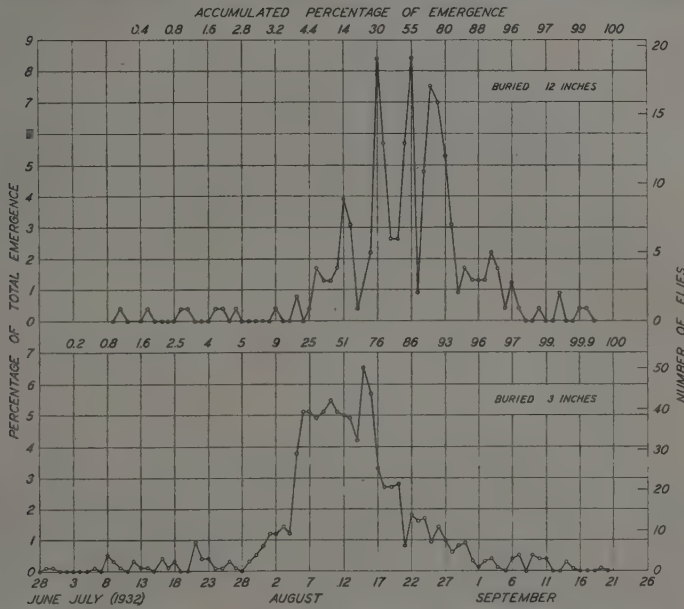



Fig. 48. Comparison of emergence of *Rhagoletis completa* from 3-inch and 12-inch depths in the soil in 1932.

in depth. The accumulated emergence in the former on August 12 was 51 per cent, while in the latter it was 14 per cent. Furthermore, emergence was considerably more erratic in the latter .

*Mechanical Effect of Depth in Soil Upon Emergence.*—The depth at which pupae are located in the soil is related to emergence. The temperature relations to emergence resulting from depth in soil have been briefly discussed. Limited information regarding the mechanical relation was obtained. The following experiments were conducted to determine the depth from which flies are capable of emerging in normally packed soil and in unpacked or relatively loose soil. Early in the season 200 live puparia were placed in soil in the bottom of each of ten 16-mesh

galvanized-wire-screen cages. These cages were 7 inches in diameter and the walls and bottoms were lined with wax paper to keep the emerging adults within the confines of the cage. All soil used was taken from the infested grove in which the experiment was conducted, and was sifted before being placed in the cages. In series *A* the soil was packed in a uniform manner to simulate orchard conditions, while in series *B* it was merely put into the cages without any effort to pack it. One *A* cage and

TABLE 2

DEPTH FROM WHICH ADULTS MAY EMERGE IN PACKED AND UNPACKED SOIL

| Cage No. | Depth of puparia, in inches | Total number puparia buried | Empty puparia | | Number live pupae at end of season | Number puparia unaccounted for | Flies collected | |
|---|---|---|---|---|---|---|---|---|
| | | | Number | Per cent of total | | | Number | Per cent of total that emerged* |
| | | | | Packed soil | | | | |
| A1 | 8 | 200 | 137 | 69 | 40 | 23 | 26 | 19 |
| A2 | 14 | 200 | 131 | 66 | 41 | 28 | 20 | 15 |
| A3 | 20 | 200 | 122 | 61 | 60 | 18 | 21 | 17 |
| A4 | 26 | 200 | 128 | 64 | 43 | 29 | 13 | 10 |
| A5 | 34 | 200 | 61 | 31 | 52 | 87 | 1 | 2 |
| | | | | Unpacked soil | | | | |
| B1 | 8 | 200 | 135 | 68 | 36 | 29 | 48 | 36 |
| B2 | 14 | 200 | 132 | 66 | 31 | 37 | 10 | 8 |
| B3 | 20 | 200 | 115 | 58 | 38 | 47 | 72 | 63 |
| B4 | 26 | 200 | 131 | 66 | 49 | 20 | 44 | 34 |
| B5 | 34 | 200 | 98 | 49 | 50 | 52 | 20 | 20 |

* As indicated by empty puparia.

one *B* cage were placed at each of five depths in soil in a walnut grove so that the pupae were 8, 14, 20, 26, and 34 inches below the surface. Adults emerging on the surface in these cages during the season were collected at regular intervals. At the end of the season the soil in each cage was carefully sifted to obtain the empty pupal cases and the puparia from which no emergence had taken place. The results of these experiments are presented in table 2.

The data show that packing the soil materially affected the ability of emerging flies to reach the surface from all depths. It is of particular interest to note that 1 fly in the packed soil and 20 flies in the unpacked soil reached the surface from the depth of 34 inches. The percentage of total emergence was considerably lower at the 34-inch depth than at other depths, while the mortality was appreciably higher. Soil tempera-

ture at this depth may have affected the percentage that emerged. The erratic nature of certain of the data is difficult to explain. With cage *B–2*, where puparia were at a depth of 14 inches, only 8 per cent of those that emerged were collected on the surface; while with cages *B–3* and *B–4*, with puparia at 20 and 26-inch depths, 63 and 34 per cent, respectively, were collected. It is possible that the cages may have been invaded by predacious insects such as certain ground beetles or ants. However, at no time was there any indication of such insects having been present.

The number of puparia unaccounted for is also difficult to explain. It is probable that the pupae died shortly after the experiment was set up, in which case the pupal shell may have deteriorated sufficiently for broken parts to pass through the regular sifting screen. Since there were considerable numbers missing in each cage, it is indicative that similar factors were operating in all cases.

*Time of Day of Emergence.*—Limited studies were conducted to determine the time of day at which emergence took place. This was of considerable importance with reference to the time of day at which collection of flies from emergence cages could be most advantageously made. Information was obtained by collecting the flies every two hours from a few representative cages. These data are presented in table 3.

TABLE 3

SUMMARY OF TWO-HOUR OBSERVATIONS REGARDING EMERGENCE FROM SOIL ON AUGUST 18, 19, 20, 1929

| Hour | Mean air temperature, degrees F | Mean humidity, in per cent | Number of flies emerging | | | Per cent of total daily emergence | |
|---|---|---|---|---|---|---|---|
| | | | Male | Female | Total | Mean | Cumulative |
| Morning | | | | | | | |
| Before 6 | 62 | 86 | 8 | 4 | 12 | 3 | 3 |
| 6– 8 | 76 | 66 | 79 | 91 | 170 | 39 | 42 |
| 8–10 | 83 | 43 | 85 | 113 | 198 | 43 | 85 |
| 10–12 | 94 | 38 | 21 | 22 | 43 | 10 | 95 |
| Afternoon | | | | | | | |
| 12–2 | 95 | 37 | 4 | 5 | 9 | 2 | 98 |
| 2–4 | 88 | 42 | 1 | 2 | 3 | 1 | 99 |
| 4–6 | 80 | 54 | 2 | 3 | 5 | 1 | 100 |

It is evident that the major portion of the daily emergence occurred in the forenoon before ten o'clock. Apparently this feature of emergence is related to temperature. In this instance it is distinctly advantageous to the species to emerge before soil-surface temperatures be-

come high enough to cause mortality. On several occasions when newly emerged flies were placed on the soil surface at the peak of the day's temperature, death quickly ensued. On days of relatively cool periods it was commonly observed that emergence was fairly uniform in rate throughout most of the day.

*Discussion of Emergence Data.*—A partial summary of the emergence data for the five-year period is presented in table 4. It is particularly interesting to note that biennial-generation individuals represent a relatively high percentage of total population and furthermore that a portion does not emerge until the third and fourth years after pupation. There were no second-generation individuals in either 1928 or 1930, when these experiments were conducted.

These studies show important relations of temperature to emergence. The data already presented suggest that the time of initial emergence is dependent somewhat upon accumulated temperature effects during dormancy. The winter seasons of 1927–28, 1928–29, and 1931–32, are classed as cold, that is, with nearly normal or subnormally low temperatures, while those of 1929–30 and 1930–31 are classed as mild, that is, with abnormally high temperatures. The medians of emergence in the former instances were reached on August 17, 24, and 16, respectively; while in the latter instances the medians were comparatively early, occurring on July 29 and 18, respectively. In this connection it is of interest to note that the percentage of annual-generation flies after so-called cold winters was approximately 60 per cent, in contrast to approximately 86 per cent after mild winters. Furthermore, after the cold winters females were more abundant than males during the early portion of the emergence season, while males predominated during the latter portion. Also the average sex ratio for these three years shows slightly more females, while the situation is reversed in 1930 and 1931. After mild winters the two sexes emerged at about the same rate.

The rate of emergence is apparently related to current soil temperature, which is directly related to air temperature. Soil temperature fluctuations usually lag 2 or 3 days after fluctuations of air temperature. After emergence has been initiated the rate is low when the mean daily air temperature is 65° F or below; however, at no time during the emergence period was the mean below 60° F except late in the season when most of the flies had emerged. At 70° F the rate increases and is rapidly accelerated by higher temperatures.

The data comparing emergence of annual and biennial-generation flies are summarized in table 5. Biennial-generation flies reached the median of emergence 7 days earlier on the average than did the annual-generation ones. Accumulated temperature effects during dormancy

TABLE 4

EMERGENCE OF WALNUT HUSK FLY, 1928–1932, SHOWING SEX RATIO AND DISTRIBUTION OF GENERATIONS

| Year | Date when seasonal median reached | Total number of cages | Number of flies | | | Ratio ♀ : ♂ | Number of healthy pupae at end of season | Approximate percentage emerged after pupae remained in soil for period of:* | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | ♀ | ♂ | Total | | | 1 year | 2 years | 3 years | 4 years |
| 1928 | August 17 | 4 | 759 | 700 | 1,459 | 52 : 48 | 640 | 69.9 | 28.7 | 1.2 | 0.2 |
| 1929 | August 24 | 20 | 3,956 | 3,442 | 7,398 | 53 : 47 | 9,203 | 44.6 | 55.1— | 0.3 | .... † |
| 1930 | July 29 | 15 | 4,318 | 4,647 | 8,965 | 48 : 52 | 1,014 | 89.8 | 9.3— | 0.9 | .... † |
| 1931 | July 18 | 12 | 5,564 | 6,292 | 11,856 | 47 : 53 | 2,395 | 83.2 | 16.8— | .... † | .... † |
| 1932 | August 16 | 15 | 3,688 | 3,712 | 7,400 | 50 : 50 | 3,718 | 66.5 | 33.5— | .... † | .... † |
| Total | | 66 | 18,285 | 18,793 | 37,078 | 50 : 50‡ | Mean | 70.8 | 28.7— | 0.8 | 0.2 |

* Pupal mortality was not considered in calculating these data.

† Data not available.

‡ Unweighted average.

may be offered in partial explanation of the facts; however, it is probable that genetical factors exert greater influence with respect to biennial-generation individuals than does temperature.

TABLE 5

Comparison of Emergence of Annual and Biennial Generation from the Soil for a Four-Year Period

| Year | Approximate date when 50 per cent of total emergence occurred | | Difference (− = earlier, + = later) for biennial generation |
|---|---|---|---|
| | Annual generation | Biennial generation | |
| | | | *days* |
| 1929 | August 24 | August 17 | − 7 |
| 1930 | July 29 | July 31 | + 2 |
| 1931 | July 26 | July 13 | −13 |
| 1932 | August 12 | August 1 | −11 |
| Mean | ——— | ——— | − 7 |

The data comparing emergence from cages distributed with respect to tree exposure are summarized in table 6. The median of emergence was reached approximately 7 days earlier in cages on the south side of trees, and 10 days earlier in those on the west side, than in those on the north side. These differences are apparently related to soil temperature since the south and west exposures receive more sunlight than the north, which is shaded by the tree a great deal of the time.

TABLE 6

Comparison of Emergence from Soil in Cages Located on Various Exposures of Trees

| Year | Approximate date when 50 per cent of total emergence occurred | | | Difference (+ = later) | | |
|---|---|---|---|---|---|---|
| | North | South | West | North as compared with south | North as compared with west | South as compared with west |
| | | | | *days* | *days* | *days* |
| 1931 | July 26 | July 19 | July 16 | +7 | +10 | +3 |
| 1932 | August 21 | August 14 | ——— | +7 | ——— | ——— |
| Mean | ——— | ——— | ——— | +7 | +10 | +3 |

Regarding the effect of depth of burial upon emergence, the data show that the median of emergence was reached approximately 10 days earlier in the 3-inch depth cage than in the 12-inch depth one (table 7). Both cages were located identically, therefore soil temperature is suggested in explanation of the facts.

The data presented in table 7 indicate further interesting relations of temperature to emergence. It is apparent that time and rate of emergence, and also the actual number of flies that emerge from any given location in one season, are related to temperature. In 1931, which was a mild winter, there was no appreciable difference in the percentage that emerged from the north and the south locations. However, in 1932 (cold winter) 27 per cent more flies emerged from the south locations than from the north locations. Temperature during dormancy is also correlated with the percentage of individuals that constitute the annual and biennial generations respectively.

TABLE 7

EFFECT OF LOCATION IN SOIL AND AGE UPON PERCENTAGE OF TOTAL PUPAE FROM WHICH ADULTS EMERGE

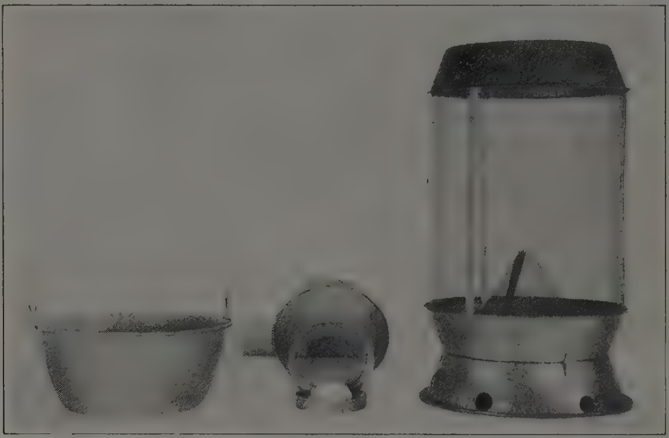
| Year | Location of cage under tree | | Age of pupae | | Depth pupae buried in soil | |
|---|---|---|---|---|---|---|
| | North | South | 1 year | 2 years | 3 inches | 12 inches |
| | Per cent of total pupae in soil from which flies emerged | | | | | |
| 1931 | 82 | 81 | 81 | 95 | .... | .... |
| 1932 | 52 | 79 | 65 | 83 | 83 | 60 |
| Mean | 67 | 80 | 73 | 89 | 83 | 60 |

*Chemotropism of the Adult.*—A considerable amount of research has been done in the field of chemotropic responses of the Trypetidae, particularly with regard to *Ceratitis capitata* Wied. (Mediterranean fruit fly), *Pterandrus rosae* (Ksh.) (Natal fruit fly), and others. However, definite information regarding the chemotropic reactions of members of the genus *Rhagoletis* is limited. In view of the great interest in this field and the possibility that information obtained might have a bearing on the control of *Rhagoletis completa,* this phase of the study received a considerable amount of attention. Olfactometer experiments employing either McIndoo's(28) or Ripley and Hepburn's(32) types of apparatus, or both, would have been highly desirable, but such time-consuming studies were not possible. The methods employed therefore were largely random testing of readily available volatile organic materials to determine whether or not there was an appreciable positive response by the flies. Rather simple and crude preliminary indoor laboratory tests, using the various chemicals, were carried out first.

All materials tested in the laboratory were subsequently given field trials. In some instances the less expensive materials were sprayed onto the foliage, but this method was obviously not suitable for the desired

studies. In other trials, pieces of cheesecloth of uniform size (1 foot square) were saturated with the material and hung in the trees.

For want of a better field method, all factors considered, the final tests were conducted in the following manner: A total of 500 flies approximately equal in sex ratio and of varying ages was liberated in each cage of a series of cheesecloth cages ($12 \times 12 \times 12$ feet) over bearing walnut trees. As indicated previously this type of cage apparently does



*A* *B* *C*

Fig. 49. Container and traps used in field chemotropic studies: *A*, open saucepan; *B* and *C*, traps. *A* and *C* are used mainly with fermenting molasses baits. *B* is used mainly in studies of various organic chemicals in large cages enclosing trees. The absorbent cotton contained in a section of petri dish, shown in the detached base, was saturated with the chemical to be tested.

not seriously affect the normal behavior of the flies. No cage was within 150 feet of another, nor in the direct path of another with reference to the prevailing winds. Two small fly traps (fig. 49 *B*) containing the chemical to be tested were hung in each caged tree, one in the lower part and the other in the upper part.

The adaptability of this type of trap was fairly well established when baited with fermenting molasses and hung in infested trees under natural conditions. A few specimens of *Rhagoletis completa* were captured, which demonstrated that *completa* was attractable and would enter traps. Fairly compact wads of absorbent cotton were saturated with the chemical to be tested and were placed in a section of a 1½ inch petri dish, which prevented direct contact between the chemical and the trap.

With the exception of a few of the very highly volatile materials, such as methyl acetate and petroleum ether, the cotton was at least moist and the odor still present at the end of 24 hours' exposure. The highly volatile materials were renewed at the end of 16 hours, before any of them was completely dissipated. Each chemical tested was exposed for 24 hours. Observations were made three times daily—in the early morning, in the midafternoon, and in the early evening before dusk—to ascertain whether or not any flies had entered the traps or were in the immediate vicinity. It is further assumed that had the material been sufficiently attractive to receive further consideration the flies would have entered the traps. Over 100 materials were tested in the above manner. All gave neutral results.

Considering the fact that the insect is practically monophagous, one would logically expect that the essential oils of the Persian walnut would produce a positive chemotropic stimulation. A small amount of walnut oil was obtained through destructive distillation of leaves and husks. Experience showed that the leaves were more readily handled and seemed to yield more oil, though neither tissue was rich in oil. The oil was tested thoroughly in the laboratory and field with neutral results.

The materials tested are as follows:

*Acids*

Acetic
Benzoic
Butyric
Naphthenic
Salicylic
Tannic

*Aldehydes*

Benzaldehyde
Butalyde
Cinnamaldehyde
Formaldehyde
Furfural
Salicylaldehyde

*Alcohols*

Amylic
Benzol
Butyl
Iso-butyl
Caprylic
Diacetone
Ethyl
Glycerol
Methanol
Methanol (synthetic)
Iso-propyl
n-Propyl
Tulol

*Esters*

Amyl acetate
Butyl acetate
n-Butyl propionate
Di-butyl phthalate
Ethyl acetate
Ethyl lactate
Ethyl sulfate
Iso-amyl benzoate
Methyl salicylate
Methyl-acetate

*Hydrocarbons*

Ethyl benzyl
Naphthalene
Toluene
Xylene

*Substituted hydrocarbons*

Bromobenzene
Chlorobenzene
Ethylene dichloride
Nitrobenzene
O-nitrochlorobenzene

*Mineral compounds*

Creosote
Distillate
Eocene
Gasoline

*Mineral compounds (continued)*

Kerosene
Petrolatum amber
Petroleum ether
Red engine oil
Tar oil

*Phenols*

Cresol
Phenol
Resorcinol

*Amines*

Ethyl benzol aniline
O-toluidine
Triethanolamine

*Terpenes*

Borneol
Camphor
Citronellol
Cymene
Eugenol
Geraniol
Terpineol

*Essential oils*

Oil aniline
Oil caraway
Oil cedarwood
Oil chamomile coctum

*Essential oils (continued)*

Oil citronella
Oil cloves
Oil eucalyptus
Oil fennell
Oil guajacwood
Oil lavender
Oil lemon
Oil linseed
Oil mustard
Oil origanum
Oil peppermint
Oil pine
Oil red thyme
Oil sassafras
Oil star anise
Oil sweet almond
Oil sweet orange
Turpentine
Oil thuja
Vanilla extract
Oil walnut (Eureka variety)

*Miscellaneous*

Ammonium hydroxide
Clensel (proprietary soap)
Distilled water
Fish oil
Pyridine
Sulfur
Quassia chips

Certain bait experiments were carried out in which ½-gallon dark-enameled, open saucepans with bails were employed (fig. 49 *A*) as traps. The following materials were tested: shredded walnut husks, shredded walnut leaves, essential oil of walnut, cane-sugar solution in concentrations of 5, 10, 25, and 50 per cent, and also the proprietary compound "Clensel," which has given such striking results in attracting large numbers of the Mediterranean fruit fly in Australia. The saucepans containing the various materials in water were hung in the lower portions of the trees. Six trees in a heavily infested walnut grove were employed in testing each material. The experiments extended over a period of twenty days at the height of fly activity, with neutral results. An occasional fly of the *completa* species was captured in the liquid in the bait pans, but this was probably an accidental occurrence since they were also found occasionally in the controls which contained water.

Preliminary tests were also conducted with fermenting baits. Frost[16] and Peterson[29] have studied this subject in great detail with reference to the moth *Laspeyresia molesta* Busck (Oriental peach moth). Frost reports in his *Laspeyresia* studies that great numbers of *Rhagoletis cingulata* Loew (light-banded cherry fruit fly) were taken in bait pans containing fermenting molasses. Ripley and Hepburn[33] in South Africa

made detailed studies of fermenting baits in relation to the trypetid *Pterandrus rosae* (Natal fruit fly). They worked primarily with treacle in combination with pollard and bran.

In these studies dealing with *Rhagoletis completa* the various materials were hung in the trees in open saucepans. The experiments were conducted in a fairly heavily infested grove at the height of fly activity and extended over a period of twenty days. Twelve trees were employed

TABLE 8

ADULTS CAPTURED IN OPEN BAIT PANS CONTAINING FERMENTING SUBSTANCES

| Experiment No. | Materials | Concentrations | *Rhagoletis completa** | | | |
|---|---|---|---|---|---|---|
| | | | ♂ | ♀ | Total | Average per pan† |
| 1 | New Orleans molasses<br>Water | 50 cc<br>950 cc | 37 | 25 | 62 | 5.2 |
| 1*a* | New Orleans molasses<br>Water<br>Sodium arsenite | 50 cc<br>950 cc<br>1 cc | 24 | 15 | 39 | 3.3 |
| 2 | Wheat bran<br>New Orleans molasses<br>Water | 90 grams<br>20 cc<br>1 liter | 32 | 19 | 51 | 4 3 |
| 3 | Middlings (Pollard)<br>New Orleans molasses<br>Water | 90 grams<br>22 cc<br>1 liter | 56 | 28 | 84 | 7.0 |
| 4 | Control (water) | ........ | 5 | 2 | 7 | 0 6 |

* Among other species of insects captured in these bait pans were noctuids, coccinellids, chrysopids, drosophilids, sapromyzids, ortalids, and other miscellaneous species.

† 12 pans in each experiment.

in testing each particular material or combination of materials. One container was hung in each tree. The materials used, concentrations, and results, are summarized in table 8.

The materials and concentrations used in experiments 1 and 1*a* of table 8 were also employed in screen fly traps (fig. 49 *C*) hung in the trees. The results obtained were essentially in accordance with those reported for the respective materials in table 8.

The data of table 8 indicate that *Rhagoletis completa* is positively chemotropic to volatile products of fermentation. The attractiveness is apparently selective with regard to sex since more males were consistently captured than females. However, the differences may not be significant as the fly population in this particular grove at that time may have consisted of an excess of males over females. The addition of sodium arsenite to the molasses-water bait appreciably reduced the rate of

fermentation, which fact in turn apparently reduced the attractiveness of the material. Other workers have reported similar results in tests with other insects.

It is recognized that the studies on chemotropic responses herein reported were not sufficient to base important conclusions upon. However, they indicate that this line of attack does not merit extensive investigation in connection with control.

*Phototropism of the Adult.*—Limited experiments were conducted relating to the phototropic responses of the flies. Laboratory tests, in which regular Mazda electric light bulbs of clear, yellow, red, green, and blue were employed, resulted in inconclusive information. In the field tests, lights of these colors were suspended in wire-screen cages enclosing walnut trees, and hundreds of flies were liberated in the cages. Neutral results were obtained.

Small-scale field tests were conducted in an effort to determine whether or not flies would exhibit a positive response to white materials on walnut foliage. The following white materials were employed: talc, hydrated lime, arsenate of lead, and barium fluosilicate. Each of these white materials was used singly and in contrast with the black material, lampblack (carbon). Lampblack was also used singly. The end of one branch in a favorable location on each of four walnut trees in a moderately infested grove was treated until no more material would adhere to the foliage. Where the black and white materials were contrasted, a similar area in close proximity on the same tree was treated likewise with the contrasting material. These treated areas were closely observed twice daily for a period of one week in order to compare the numbers of flies present on each. The results are considered to be neutral, since the flies exhibited no particular response by frequenting treated foliage; neither were they repelled.

*Anemotropism and Thermotropism of the Adult.*—From laboratory anemotropic studies it was concluded that flies respond neutrally to this stimulus, except under extreme conditions wherein they are forced to orientate themselves with head in the direction of air current for protection. Simple thermotropic tests within the temperature range of 70° to 130° F showed that the flies were neither attracted nor repelled by temperatures that were not detrimental to them. However, when the temperature of their environment was above 120° F, they became excitedly active. At higher temperatures they succumbed with relatively short exposures.

*Geotropism and Thigmotropism of the Adult.*—On emerging from puparia in the soil, the adults indicate strong negative geotropism. The

path made by newly emerged adults has been observed in detail. Puparia placed against the surface of glass containers, and covered with very finely divided pure sand to a depth of 6 to 8 inches, afford a good method of studying the path made by the emerging adult, provided the sand is thoroughly wetted to a depth below the location of the puparia. In every instance in the many cases observed the path has always been upward though not necessarily vertical. Under field conditions the emerging adults no doubt follow the path of least resistance, taking advantage of horizontal cracks that eventually lead to cracks extending upward. The indications are that the flies do not respond to geotropic stimuli after they have reached the surface and their bodies become hardened.

A positive thigmotropism may be indicated by the behavior of the adults immediately upon emerging from the puparium. Dead bodies of newly emerged flies were often found in the center of tightly rolled cotton plugs in glass test tubes where puparia were contained without soil. Some of the test tubes were upright and some horizontal in position and each contained only small numbers of puparia in the bottoms of the tubes. Therefore space was not particularly limited. In those instances where the test tubes were upright these observations may simply indicate a negative geotropic response; however, where the tubes were horizontal positive thigmotropism is suggested.

*Feeding Habits.*—The food of adults under natural conditions is not definitely known. Illingworth[23] in his studies on *Rhagoletis pomonella* (Walsh) on apples states, "The surface gum [on the fruit] is apparently the only food taken when the flies are in the orchard, for when they go to the leaves it appears to be for rest and for shelter from the weather." It seems improbable that gum or wax alone would supply sufficient nutritive substances, if any at all.

Honeydew resulting from aphid infestation undoubtedly constitutes a portion of the food of *R. completa*. The walnut aphid, *Chromaphis juglandicola* (Kalt.) is always present in greater or lesser numbers throughout the period that the tree is in foliage. Sufficient numbers are usually present in the early portion of the season to excrete enough honeydew for visible amounts of sooty mold fungi to develop. Consequently quantities of honeydew and spores of fungi are available for food throughout the period of fly activity.

Yeasts occurring naturally on the foliage and bark are probably ingested and serve a nutritive purpose. Evidence of ingestion is forthcoming from Berlese's work (Caldis[9]). In his studies of souring of figs, he found that yeasts multiplied and also hibernated in the intestines of a number of species of flies. Furthermore the surface of the walnut bears

small glandular hairs. It is not improbable that the cells of these glands are readily ruptured by the fly. The contents of such cells should be rich in nutritive substances. Plant sap oozing at pruning scars and also from lesions due to invading bacterial and fungal organisms is also consumed by the flies.

Feeding may take place at any time of day; however, the flies generally feed more actively in the early morning when climatic conditions are favorable. Atmospheric moisture in the form of dew is usually present until about eight o'clock on a typical morning with sunshine. They move about very little if at all at temperatures of 50° F and below and are quite sluggish even at 55° F. However, when temperature reaches approximately 60° F after daylight they begin to move about, apparently in search of food. This movement consists of walking about accompanied by short flights. The rate at which they walk about increases as the temperature rises and apparently is also stimulated by the rapid evaporation of dew. It thus seems that the presence of moisture materially aids them in the ingestion of food; however, it may mechanically hinder locomotion. Extensive field observations indicate that feeding takes place at a time of day when flies are likely to obtain food with the least effort.

In feeding, the flies walk about on the leaves and nuts with proboscis extended downward, sampling the surface for the presence of food. When a favorable spot is located, a rapidly pulsating action takes place at the base of the mouth parts, which probably indicates that some material is being sucked up. The proboscis is frequently cleaned with the aid of the forelegs. They consume quantities of liquid by sucking up droplets of dew. In many instances also flies have been observed to emit a small amount of fluid onto the surface, then move the fleshy labella about in a rasping manner, and later suck it up again. This no doubt serves to change the desired food material into a liquid state, or causes small solids to become suspended in the liquid for consumption. This is the usual procedure when feeding upon sweetened materials that have been applied on the foliage. When the food supply is plentiful the flies engorge until their abdomens are greatly distended and a droplet of liquid usually surrounds the labella of the proboscis. Such engorged condition appears to inhibit activity temporarily to a considerable extent.

In relation to the control of the fly, it was desirable to study the action of the mouth parts with reference to consumption of more or less insoluble particles of insecticides. These ingestion studies are reported elsewhere in this paper (p. 510) since they pertain more specifically to toxicological data.

*Flight and Dispersion.*—On casually examining flies morphologically the impression is gained that they are well adapted for strong flight. The flights are usually of short duration. The flies are very rapid in "taking off" and also while on the wing; many of their flights are begun by characteristic "darts." They have a fast "landing speed" and come to rest on the leaf with a characteristic noise or thud. In field studies it commonly happened that flies were located very readily by the sound made on alighting on a nearby leaf.

With reference to the general migration of flies from tree to tree within an infested grove, little is definitely known. Since a satisfactory attrahent with which traps may be baited has not been found, it is difficult to obtain detailed information regarding dispersion. Flies have been observed to travel on the wing from one tree to another. Detailed counts of the degree of infestation on all trees within a grove show that the infestation within a unit area is not homogeneous. This fact may be explained by a tendency for flies to localize on the tree that they first reached on emerging from the soil. Another possible explanation would be that, because of the hardness of walnut husks of certain individual trees, the females were unable to oviposit and began to migrate in search of susceptible walnuts. The trees bearing nuts with susceptible husks were therefore located by the flies as a result of "trial and error."

In migration studies in 1929, two separated rows in a 10-acre ($660 \times 660$ feet) block of Elberta peaches were sprayed with sugar solution. Flies were observed on all sprayed trees. This peach planting adjoined a 10-acre block of Eureka walnut trees that were heavily infested in 1928. Since the peaches were harvested before complete maturity was reached, very few of the flies present could have developed on this host. Therefore most of the adults in this peach planting either migrated there voluntarily or were subjected to a dispersing effect by the prevailing southwest winds.

An effort was made in 1930 to study the movement of flies within a small, infested Eureka walnut grove. For this purpose flies were permanently marked. Several methods were tested, namely, clipping a portion of the wing, marking the wing with India ink, amputating one leg, and other methods, none of which was satisfactory for field studies. The marking of the scutellum with India ink, however, proved satisfactory, since it did not injure the fly and was easy to detect. The scutellum is normally yellowish white and is very conspicuous, because of the contrast afforded by the darker colored body. Of the various colors tested either brick red or light green was satisfactory, though brick red was slightly more conspicuous and was used for this reason. To accomplish the marking, the flies were placed in short, wide-mouthed fruit jars, 25

individuals per jar, and these were then kept at a temperature of approximately 37° F for one-half hour. This treatment inactivated the flies, and under these conditions a small drop of the India ink was placed on the scutellum with a camel's hair brush. Four hundred flies were marked in this manner in approximately one hour. Of these, 350 flies were liberated on one medium-sized walnut tree. The remaining 50 flies were placed in the inverted battery-jar cage on walnuts in the field laboratory to study the effect of the treatment on the flies. They remained alive and behaved normally throughout the remainder of the season. The scutellum marking was permanent and was very conspicuous.

Sugar solution was sprayed onto the tips of branches in the lower, middle, and upper portions of each of the four sides of the tree on which the flies were liberated. The same solution was also placed on the lower portion on four sides of all peach and walnut trees within a radius of 100 feet. Daily observations were made in order to note the number of marked flies feeding on the artificial food in the various locations of the tree on which they were liberated; and also to note whether or not any had migrated to surrounding trees. The greatest number of marked flies observed in any one day was 12. These were feeding on the sugar in the top of the tree on the southeast portion during late afternoon. Occasionally a single marked fly was noted feeding in other portions of this tree. In no instance was a marked fly recorded from any of the surrounding trees. For studies of this nature a large number of marked flies, perhaps 8,000 to 10,000, would be necessary to yield sufficient data to be representative of normal dispersion.

In 1932, Phipps and Dirks,[30] studying dispersion of *Rhagoletis pomonella,* reported important experiments in which marked flies were used. Their data show that 27 per cent of the marked individuals that were recovered were taken within a radius of 75 yards; 57 per cent between 75 and 96 yards; and 16 per cent from 98 to 156 yards.

Yearly increase of infested acreage, as an index to normal dispersion, supplies suggestive data. However, an extensive control campaign has been conducted since 1928, which may have affected normal dispersion somewhat. Yearly surveys of the same area showed increases in infestation amounting to from ¼ to 3 miles, in a straight line, per year.

*Population Studies.*—For purposes of control, information regarding the relative numbers of flies present on trees in various areas was highly important. In 1928 it was observed that sweetened materials applied on the foliage served to congregate the flies. Cane sugar was most commonly used in preliminary studies. Since this substance possessed no chemotropic attraction, the fact that the flies became congregated in such

small areas as on a few leaves on the tip of a branch indicates that they moved about generally over the trees.

For these studies a grove of medium-sized trees was chosen, in which a fairly high fly population was known to exist since many walnuts showed evidence of recent infestation. On each of two adjoining trees a 20 per cent cane sugar solution was sprayed onto leaves at the tip of a branch on the north, east, south, and west locations. On three other trees in an adjacent row the material was applied only on the east and west locations. Detailed observations and counts of flies present were made at hourly intervals from 6:00 a.m. until 6:00 p.m. on two consecutive days. Early in the morning of the third day, canvas was placed under each tree and the trees were heavily dusted with high-concentration nicotine dust. Application was made under favorable conditions and before the flies had begun to move about. The indications were that practically all flies were brought down onto the canvas. The flies were collected before they revived. On the following morning the trees were again dusted and a few flies were brought down. Five days later sugar solution was again sprayed onto the same areas on the same trees and also in similar locations on other trees within the grove. Very few flies were noted on the former trees, while flies were fairly abundant on the latter. These facts indicate that the flies do not move about much from one tree to another, provided food and host conditions favoring oviposition prevail. This assumption is made in interpreting the data regarding this phase of the study.

The data obtained from these studies are presented in table 9. Considerable variation in the behavior of the flies is evident. On September 5, over twice as many flies were observed on the north and east locations as on the south and west sides of the trees. Furthermore, when only the east and west locations were observed, many more flies congregated on the easterly side. It is of interest to note that on this morning atmospheric moisture did not condense in sufficient amounts to form dew, and the sun shone brightly all day.

On September 6, a different picture is presented. A high fog prevailed throughout most of the forenoon and during the remainder of the day the sky was overcast; the sun did not shine brightly at any time. This condition apparently affected the normal habits of the flies, particularly with respect to their movement about the trees. Less than one-half as many flies were observed as on the preceding day, and there were no significant differences in the numbers frequenting any of the respective locations except in the southerly location.

On both days appreciably more flies were observed in the afternoon than in the forenoon. A summary of the composite data secured is

TABLE 9

Results of Observations on Two Consecutive Days Concerning the Activity and Distribution of Adults on Walnut Trees (1930)

| Hour | Temperature, degrees F | Relative humidity, in per cent | Two trees with total population of 146 flies | | | | | Three trees with total population of 230 flies | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | North | East | South | West | Total | East | West | Total |
| | | | Flies observed, in per cent of daily total* | | | | | | | |
| September 5 | | | | | | | | | | |
| 6 a.m. | 56 | 56 | 0 | 0 | 0 | 0 | 0 | 1 | 0 | 1 |
| 7 a.m. | 63 | 44 | 1 | 0 | 3 | 1 | 5 | 0 | 0 | 0 |
| 8 a.m. | 75 | 41 | 1 | 3 | 1 | 1 | 5 | 0 | 0 | 0 |
| 9 a.m. | 84 | 35 | 4 | 1 | 1 | 1 | 8 | 2 | 1 | 3 |
| 10 a.m. | 90 | 35 | 6 | 7 | 1 | 5 | 19 | 5 | 6 | 11 |
| 11 a.m. | 92 | 35 | 4 | 7 | 1 | 7 | 19 | 6 | 4 | 10 |
| 12 a.m. | 94 | 33 | 9 | 9 | 1 | 2 | 21 | 7 | 3 | 10 |
| Total a.m.* | *M 79* | *M 40* | *25* | *27* | *9* | *16* | *77* | *19* | *14* | *35* |
| 1 p.m. | 95 | 35 | 5 | 8 | 3 | 5 | 22 | 6 | 3 | 10 |
| 2 p.m. | 93 | 36 | 9 | 14 | 1 | 4 | 30 | 8 | 6 | 14 |
| 3 p.m. | 92 | 40 | 6 | 10 | 3 | 3 | 21 | 9 | 3 | 12 |
| 4 p.m. | 91 | 42 | 5 | 10 | 3 | 5 | 23 | 7 | 4 | 11 |
| 5 p.m. | 85 | 47 | 7 | 8 | 3 | 4 | 23 | 6 | 3 | 9 |
| 6 p.m. | 77 | 58 | 7 | 3 | 1 | 1 | 13 | 5 | 4 | 9 |
| Total p.m.* | *M 89* | *M 43* | *38* | *54* | *14* | *23* | *130* | *41* | *23* | *64* |
| Daily total* | *M 84* | *M 41* | *64* | *81* | *23* | *39* | *208* | *62* | *37* | *99* |
| September 6 | | | | | | | | | | |
| 6 a.m. | 55 | 63 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| 7 a.m. | 60 | 58 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| 8 a.m. | 69 | 51 | 0 | 2 | 0 | 0 | 2 | 1 | 0 | 1 |
| 9 a.m. | 75 | 42 | 1 | 2 | 3 | 0 | 5 | 1 | 1 | 2 |
| 10 a.m. | 83 | 47 | 5 | 3 | 2 | 0 | 11 | 1 | 1 | 3 |
| 11 a.m. | 86 | 46 | 3 | 3 | 1 | 3 | 10 | 2 | 1 | 3 |
| 12 a.m. | 87 | 49 | 3 | 1 | 0 | 2 | 7 | 2 | 3 | 5 |
| Total a.m.* | *M 74* | *M 51* | *12* | *12* | *7* | *5* | *36* | *7* | *6* | *13* |
| 1 p.m. | 85 | 47 | 1 | 3 | 1 | 6 | 12 | 2 | 1 | 3 |
| 2 p.m. | 83 | 49 | 2 | 3 | 0 | 4 | 9 | 2 | 2 | 4 |
| 3 p.m. | 82 | 55 | 2 | 5 | 2 | 8 | 16 | 2 | 1 | 3 |
| 4 p.m. | 78 | 73 | 3 | 3 | 1 | 4 | 12 | 1 | 1 | 2 |
| 5 p.m. | 71 | 78 | 3 | 1 | 1 | 1 | 5 | 1 | 3 | 3 |
| 6 p.m. | 65 | 82 | 2 | 0 | 1 | 1 | 3 | 0 | 1 | 0 |
| Total p.m.* | *M 77* | *M 64* | *14* | *16* | *5* | *23* | *58* | *7* | *9* | *17* |
| Daily total* | *M 75* | *M 57* | *26* | *27* | *12* | *28* | *94* | *14* | *15* | *30* |

* Totals include those flies that may have been observed more than once during the interim, consequently exceeds 100 per cent in some instances.

graphically presented in figure 50. The number of flies observed on both days increased with rising temperature, reaching the daily peak at 2 p.m. and 3 p.m., respectively, and decreased with lowering temperature. The inverse relation generally existed with regard to relative humidity. The data indicate that absence of dew in early morning, followed by favorable temperature and sunshine, stimulated movement

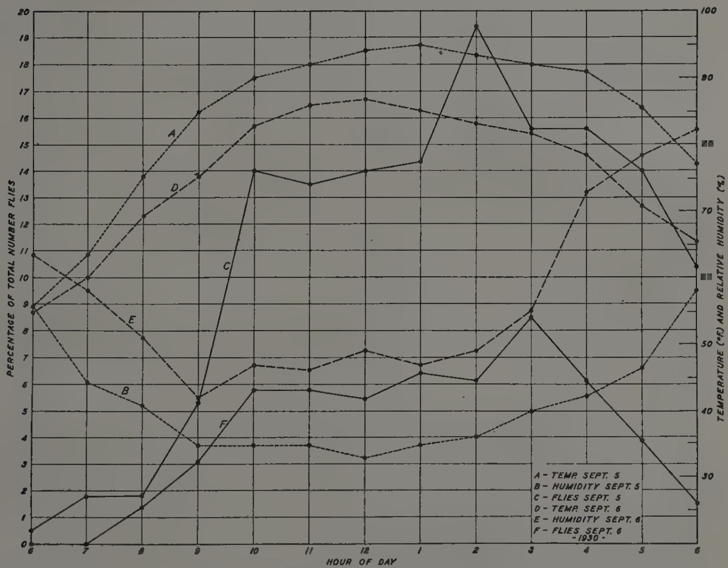



Fig. 50. Comparison of relative numbers of flies observed on the same walnut trees on two consecutive days, together with prevailing temperature and humidity (1930).

incident to acquiring food throughout most of the day. When dew was present and the temperature was favorable, movement was stimulated somewhat, though lack of sunshine apparently was related to the extent or degree of movement. However, it may be argued that sunshine had no connection with the matter and that lack of detectable movement may have been due to the fact that the flies had acquired the desired food and consequently had no need of moving about extensively.

For comparing relative populations in experimental control plots in 1929 and 1930, sugar solution was applied on a small area of foliage on the east side of representative trees. The numbers of flies observed served as an index to the population in any given plot. These studies were made during August, and on days when an appreciable quantity of dew was

present at sunrise. The temperature rose rapidly and as a result the dew evaporated quickly. Under these conditions greater numbers of flies could be observed over a period of several hours in early morning than at any other time of the day. This method was used extensively in 1929 and 1930. Since the data pertain specifically to control studies, they are treated under that phase of the subject (pp. 515–535).

*Longevity of Adult.*—Under field laboratory conditions, in the battery-jar type of cage, a few flies were kept alive in 1930 from July 22 to October 15, a total of 85 days. The average length of life under the most favorable laboratory conditions known was approximately 40 days. Virgin females lived on the average approximately 35 days, and the oldest one on record was 85 days old.

In cheesecloth cages ($12 \times 12 \times 12$ feet) enclosing small walnut trees, a few adults were alive at the end of 70 days, when the cages were removed.

The length of life under natural field conditions is not known. However, it seems reasonable to believe that an average span of life of from 30 to 40 days would be approximately normal. This is based on the assumption that the foregoing artificial conditions pertaining to longevity are probably as favorable on the whole as field conditions, or slightly more so.

*Effect of Food on Longevity.*—A simple experiment was conducted to determine the length of life of flies without food or water. One hundred flies of approximately equal sex ratio were collected immediately upon emergence from the soil. Equal numbers of these were placed in each of two inverted battery-jar cages. Approximately 50 per cent of the flies were dead at the end of 48 hours, while over 90 per cent were dead at the end of 52 hours. A few flies lived nearly four days without food.

In earlier studies granulated cane sugar, and later cube cane sugar, was put into one section of a small petri dish in battery-jar cages for food. Absorbent cotton saturated with water was put into another section of petri dish to supply moisture. Granulated sugar deliquesced and became sticky much more rapidly than cube sugar. Any sticky substance within the cage was undesirable, since the flies became trapped or, in walking about, spread the material throughout the cage to such an extent that they were frequently incapacitated when their wings came into contact with it. Therefore sticky food substances constituted a hazard which was eliminated by more frequent feeding, following the method to be described in the nutritional studies. In these studies flies lived longest under battery-jar-cage conditions in an outdoor shaded laboratory when a solution of 10 per cent technical sucrose was supplied as food. However, only a few eggs were produced on this diet. Moisture

is very essential in the economy of the fly, both for food and for maintenance of optimum humidity conditions; which role is of most importance is not entirely clear.

*Effect of Temperature on Longevity.*—The temperatures that normally prevail during the period of fly activity rarely drop lower than 40° F, and are not low enough to exert a deleterious effect on longevity. In fact, general observations over a four-year period in the laboratory and field indicate that length of life during the later (cooler) portion of the season appreciably exceeds that of the fore and middle portions. Under laboratory conditions, daily maximum temperatures around 95° to 100° F shortened life materially; and at 105° F for several hours, death ensued unless a high relative humidity was maintained. The highest temperature recorded during these studies was 114° F for nearly two hours' duration. The flies died in all of the cages except those where abundant moisture prevailed; in the latter the flies remained inactive while clustered on the saturated wad of absorbent cotton in the cage. In cheesecloth emergence cages the newly emerged flies generally left the walls of the cage and rested on shaded soil when the air temperature exceeded 100° F for half an hour or longer. At slightly higher temperatures, in emergence cages, many of the flies became inactivated and later died; however, a few revived before nightfall.

*Effect of Relative Humidity on Longevity.*—Repeated experiments under identical conditions early in this study demonstrated the superiority of the inverted battery-jar cage over a screen cage of similar size and shape. The nature of this difference appeared to be related to high relative humidity. Accordingly a comparison was made of temperature and humidity conditions existing during a period of six days in these two types of cages, together with those existing within the surrounding walnut grove. These records are presented in table 10.

It is evident from these data that the battery-jar cage maintained a higher relative humidity with only slight fluctuations. Also the temperature was somewhat reduced. The differences shown for the screen cage and walnut-grove conditions are slight and are no doubt the result of location, since the field laboratory was situated in the shade of a large walnut tree.

Further evidence on the role of humidity in the economy of the fly is forthcoming from other sources. As previously reported, flies lived for relatively long periods of time in cages ($12 \times 12 \times 12$ feet) over small walnut trees when the cages were covered with cheesecloth. However, under identical conditions except that the cages were covered with 16-mesh wire screen, most of the flies were dead in 2 or 3 weeks' time. The cheesecloth covering apparently served to maintain a favorably high

relative humidity in the immediate environs of the tree. Field observations indicate that small walnut trees do not afford environmental conditions conducive to longevity. A suggested explanation is that the relative humidity within the inner foliage is appreciably lower during the warmer portions of the day than in the case of a medium or large-sized tree under identical atmospheric conditions. The relative amounts of moisture released in transpiration from small trees and large trees may effect such differences in this semiarid region.

TABLE 10

COMPARISON OF TEMPERATURE AND HUMIDITY IN BATTERY-JAR CAGES AND IN WIRE-SCREEN CAGES, WITH NORMAL WALNUT-GROVE CONDITIONS; 1931

| Day | Hour | Temperature | | | Relative humidity | | |
|---|---|---|---|---|---|---|---|
| | | Battery-jar cage* | Screen cage* | Walnut grove† | Battery-jar cage* | Screen cage* | Walnut grove† |
| | | *degrees F* | *degrees F* | *degrees F* | *per cent* | *per cent* | *per cent* |
| July 25 | 2 p.m. | 102 | 104 | 103 | 93 | 38 | 39 |
| | 5 p.m. | 90 | 90 | 88 | 97 | 50 | 51 |
| July 26 | 9 a.m. | 82 | 85 | 89 | 98 | 74 | 58 |
| | 1 p.m. | 95 | 97 | 101 | 92 | 45 | 47 |
| | 5 p.m. | 94 | 93 | 93 | 98 | 56 | 55 |
| July 27 | 9 a.m. | 77 | 79 | 82 | 98 | 81 | 71 |
| | 1 p.m. | 96 | 97 | 98 | 92 | 51 | 52 |
| | 5 p.m. | 84 | 84 | 84 | 94 | 51 | 53 |
| July 28 | 8 a.m. | 78 | 78 | 76 | 98 | 74 | 65 |
| | 12 m. | 90 | 90 | 89 | 94 | 53 | 49 |
| | 5 p.m. | 86 | 85 | 85 | 94 | 51 | 52 |
| July 29 | 8 a.m. | 75 | 76 | 72 | 98 | 95 | 71 |
| | 12 m. | 89 | 90 | 86 | 95 | 54 | 49 |
| | 6 p.m. | 82 | 81 | 77 | 98 | 75 | 69 |
| July 30 | 8 a.m. | 72 | 73 | 69 | 98 | 95 | 77 |
| | 12 m. | 86 | 88 | 83 | 94 | 59 | 53 |
| | 4 p.m. | 85 | 84 | 82 | 92 | 64 | 58 |
| Mean‡ | All readings | 88 | 89 | 88 | 95 | 59 | 54 |

Differences between readings

| Readings compared | Temperature | Relative humidity |
|---|---|---|
| | *degrees F* | *per cent* |
| Battery-jar cage compared with screen cage | −1 | +36 |
| Battery-jar cage compared with walnut grove | 0 | +41 |
| Screen cage compared with walnut grove | +1 | + 5 |

* The battery-jar and screen cages were in a screened, shaded laboratory. Lambrecht Polymeters (mercury thermometer and hair hygrometer) were used.

† Records from hygrothermograph in standard shelter in walnut grove.

‡ Mean includes many readings during this period that were omitted from table for brevity.

Suggestive data regarding humidity were obtained in 1931. Dual-atmometer bulbs were stationed in each of the following locations: in total sunlight, 5 feet above soil surface; in center of medium-sized walnut tree, 13 feet above soil surface; and in upper portion of the same tree, 20 feet above soil surface. The evaporation of water from the surface of these bulbs throughout the major portion of the season of fly activity was obtained. These data are presented in table 11.

TABLE 11

EVAPORATION OF WATER FROM ATMOMETER BULBS LOCATED IN DEEP SHADE, PARTIAL SHADE, AND TOTAL SUNLIGHT, IN WALNUT TREE

| Bulb* No. | Total sunlight, 5 feet above soil | Deep shade, center of walnut tree, 13 feet above soil | Partial shade, upper portion of walnut tree 20 feet above soil |
|---|---|---|---|
| | Actual loss of water per bulb from August 1 to September 17 (48 days), 1931 | | |
| | *cc* | *cc* | *cc* |
| 142 | 1,616 | ........ | ........ |
| 162 | 1,516 | ........ | ........ |
| 157 | ........ | 1,240 | ........ |
| 158 | ........ | 1,016 | ........ |
| 136 | ........ | ........ | 1,771 |
| Mean loss, entire period | 1,566 | 1,128 | 1,771 |
| Mean loss per day | 33 | 24 | 37 |

* All bulbs were of S 30 series, with 0.80 correction factor.

There was an appreciable increase in the rate of evaporation of water from the atmometer bulbs located in total sunlight and in the upper portion of the tree in contrast to those located in the center of the tree. The relative humidity probably was materially higher in the inner foliage of the center of the tree than elsewhere.

*Nutritional Studies.*—In the earlier portion of the biological studies ordinary cane sugar was used as food for the flies. Other materials, such as glucose, honey, and raisins were used in preliminary studies but none seemed quite equal to sucrose in general adaptability for artificial food. However, sucrose was not entirely satisfactory, particularly with respect to fecundity of females. Fluke and Allen[(15)] in 1931 reported encouraging results with yeast suspended in honey water as artificial food for *Rhagoletis pomonella*.

The available information regarding insect nutrition (mainly based upon Uvarov[(39)]) indicates that nitrogen in the diet of adults is unnecessary for health and longevity and in fact generally shortens the life of the individual somewhat in comparison with an exclusive carbohydrate

food. An excess of proteins may be deleterious to longevity. However, nitrogen is essential for reproduction since it stimulates egg development.

Carbohydrates, aside from being necessary for longevity, have been shown to be essential for the development of genital products. The limited data indicate that sucrose and levulose are more valuable in the diet than other sugars, while glycogen and starch are of negative value without the presence of the corresponding enzymes. The role of fats is scarcely understood at all.

Little is known of the role of minerals; however, the following have been shown to affect reproduction favorably when consumed in either the larval or adult stages: potassium, phosphoric acid, magnesium, ferrous chloride, ferrous sulfate, copper sulfate, sodium hydroxide, and potash. Of the known vitamins essential in mammalian nutrition positive evidence is available regarding only one, vitamin B, as of value in insect nutrition. Yeast supplies vitamin B. However, symbiotic microörganisms may supply the necessary vitamins, in which connection practically no information is available.

In an effort to obtain data regarding the nutrition of *Rhagoletis completa,* preliminary studies were outlined and conducted in 1931. Limited time prevented a continuance of the work in 1932. There were 85 individual tests involving a total of 3,181 flies or an average of 37 flies per test. All tests were conducted in an outdoor screened laboratory located in a walnut grove in the infested area. The flies were placed in the test cages on the day that they emerged from the soil and before they took any food except negligible amounts that may have been present on the walls of the cheesecloth emergence cage. The standard method of supplying food was to saturate a wad of sterile absorbent cotton with a measured amount of the nutrient media and place it in the cage in one half of a petri dish (fig. 32, p. 402). Every second day the media was replaced with a fresh supply. Tap water was used with the technical grades of materials, while distilled water was used with chemically pure ones. All cages were thoroughly washed in tap water at weekly intervals and dried in the sun. Clusters of walnuts in susceptible condition for oviposition were thoroughly washed before being placed in the cages. These were changed at weekly intervals. Nuts removed from the cages were kept for several weeks for further studies on larval development.

It is realized that the condition of these tests, particularly with reference to control of environmental factors, was not satisfactory for precise nutritional experiments. Furthermore it was not possible to maintain an oviposition record for individual females, owing to the relatively

large number of tests involving many flies. Therefore, for comparative purposes the average number of eggs per female was calculated by dividing the total number of eggs deposited in a single test by the number of females alive when oviposition began. Thus the calculated number of eggs per female was considerably lower than the actual number. However, the data obtained should at least be indicative of the relative values of the various materials and combinations tested.

The detailed data obtained in these studies is rather voluminous and for that reason is not incorporated in this publication. Therefore, only the most important information pertaining to the experiments and a general summary of the results will be given. These data are presented in table 12.

In no instance was oviposition stimulated to the degree that is apparent under natural conditions. The maximum number of eggs recorded per female in these tests was 30. In all tests only a small number of eggs were produced per female. Further studies regarding the role of the tested materials in the nutrition and metabolism of the fly are necessary before definite conclusions are warranted. A brief summary of these studies, treated according to food groups, follows.

*Results with Proteins in Foods.*—When nitrogen was added to the carbohydrate (sucrose) diet in battery-jar cages in the form of yeast (experiments 1 and 3) the flies lived less than one-half as long and deposited only one-third as many eggs. When quartz-glass cages were used (experiment 4) a material increase in longevity and fecundity was evident in comparison with battery-jar cages (experiment 3) thus possibly indicating a slightly beneficial effect from ultraviolet or other light under these conditions. Yeast added to a honey diet in battery-jar cages (experiments 22 and 24) decreased longevity somewhat and fecundity to the extent of approximately one-third. Limited amounts of nitrogen are present in honey, mainly in the few pollen grains remaining. When glycocoll, a simple amino acid, was added to a C.P. sucrose diet (experiments 9 and 10) longevity was decreased considerably but the number of eggs deposited was doubled. Nitrogen supplied as urea (experiments 9 and 15) did not decrease longevity though fecundity was nearly doubled. When furnished as ammonia (experiments 9 and 16) longevity was not affected though fecundity was increased approximately three times. In these tests yeast apparently decreased longevity and fecundity; glycocoll decreased longevity but increased fecundity; while both urea and ammonia increased fecundity, but did not affect longevity.

*Results with Carbohydrates in Foods.*—In only a few instances (experiments 5, 8, and 14) were there appreciable differences between technical and chemically pure sucrose, under either battery-jar or

TABLE 12

SUMMARY OF DATA FROM NUTRITIONAL STUDIES

| Experiment No. | Materials used | pH of media | Type of cage | Number ♀ | Number ♂ | Eggs per ♀ | Number of days elapsed when mortality equalled approximately 50 per cent | 90 per cent |
|---|---|---|---|---|---|---|---|---|
| 1 | Sucrose (technical) 10 per cent | 6.3 | Battery jar | 78 | 100 | 3.1 | 24 | 57 |
| 2 | Sucrose (technical) 10 per cent | 6.3 | Quartz glass | 16 | 19 | 2.1 | 25 | 54 |
| 3 | Sucrose (technical) 10 per cent, yeast (dry) 3 per cent | 4.9 | Battery jar | 50 | 99 | 1.3 | 9 | 24 |
| 4 | Sucrose (technical) 10 per cent, yeast (dry) 3 per cent | 4.9 | Quartz glass | 15 | 17 | 3.2 | 19 | 35 |
| 5 | Sucrose (technical) dry | 6.6 | Battery jar | 49 | 71 | 30.0 | 5 | 9 |
| 6 | Sucrose (technical) 10 per cent | 6.3 | Screen cage hung in tree | 27 | 30 | 9.1 | 28 | 58 |
| 7 | Sucrose (technical) dry | 6.6 | Screen cage hung in tree | 86 | 138 | 0.0 | 5 | 8 |
| 8 | Sucrose (technical) 10 per cent, solution $B$* 90 per cent | 4.6 | Battery jar | 49 | 62 | 14.0 | 25 | 40 |
| 9 | Sucrose (C.P.) 10 per cent | 6.9 | Battery jar | 56 | 81 | 3.7 | 21 | 42 |
| 10 | Sucrose (C.P.) 10 per cent, glycocoll 0.5 per cent | 6.1 | Battery jar | 50 | 53 | 7.0 | 7 | 34 |
| 11 | Sucrose (C.P.) 10 per cent, glycocoll 0.5 per cent, solution $A$† 90 per cent | 5.2 | Battery jar | 32 | 46 | 14.0 | 18 | 31 |
| 12 | Sucrose (C.P.) 10 per cent, glycocoll 0.5 per cent, solution $B$* 90 per cent | 5.1 | Battery jar | 48 | 53 | 5.3 | 22 | 40 |
| 13 | Sucrose (C.P.) 10 per cent, solution $A$† 90 per cent | 4.9 | Battery jar | 39 | 42 | 9.3 | 25 | 44 |
| 14 | Sucrose (C.P.) 10 per cent, solution $B$* 90 per cent | 5.0 | Battery jar | 35 | 45 | 4.5 | 24 | 37 |
| 15 | Sucrose (C.P.) 10 per cent, urea 560 p.p.m. | 5.5 | Battery jar | 39 | 51 | 6.8 | 18 | 42 |
| 16 | Sucrose (C.P.) 10 per cent, ammonia 1,000 p.p.m. | 9.5 | Battery jar | 44 | 49 | 11.7 | 20 | 47 |
| 17 | Sucrose (C.P.) 10 per cent, levulose (C.P.) 10 per cent, dextrose (C.P.) 10 per cent, dextrin (C.P.) 10 per cent, urea 560 p.p.m. | 4.7 | Battery jar | 38 | 54 | 11.7 | 23 | 37 |
| 18 | Levulose (C.P.) 10 per cent, solution $B$* 90 per cent | 4.5 | Battery jar | 82 | 115 | 11.2 | 9 | 33 |
| 19 | Dextrose (C.P.) 10 per cent, solution $B$* 90 per cent | 3.6 | Battery jar | 54 | 83 | 5.2 | 9 | 20 |
| 20 | Dextrin (C.P.) 10 per cent, solution $B$* 90 per cent | 3.8 | Battery jar | 45 | 66 | 0.0 | 5 | 6 |
| 21 | Levulose (C.P.) 10 per cent, dextrose (C.P.) 10 per cent, dextrin (C.P.) 10 per cent, urea 560 p.p.m. | 4.4 | Battery jar | 47 | 63 | 0.0 | 3 | 5 |
| 22 | Honey 10 per cent | 4.0 | Battery jar | 115 | 133 | 7.2 | 18 | 37 |
| 23 | Honey 10 per cent | 4.0 | Quartz glass | 15 | 20 | 3.0 | 18 | 48 |
| 24 | Honey 10 per cent, yeast (dry) 3 per cent | 4.3 | Battery jar | 51 | 57 | 4.8 | 13 | 24 |
| 25 | Honey 10 per cent | 4.0 | Screen cage | 61 | 75 | 4.1 | 14 | 30 |
| 26 | Honey 10 per cent | 4.0 | Screen cage hung in tree | 52 | 55 | 16.6 | 22 | 52 |
| 27 | Water (tap) | 6.3 | Battery jar | 56 | 64 | 0.0 | 3 | 5 |
| 28 | Solution $B$* | 5.2 | Battery jar | 50 | 61 | 0.0 | 3 | 5 |

* Nutrient solution $B$ is identical to $A$, omitting Zn and Cu.

† Nutrient solution $A$ consisted of the following chemicals in parts per million: $NO_3$, 144; Mg, 11; Cl, 2; Na, 1.4; Ca, 80; K, 93; $PO_4$, 53; Mn, 0.03; $SO_4$, 72; Fe, 3; Zn, 1; Cu, 3.

quartz-glass cage conditions. In these experiments (experiments 8 and 14) mineral nutrient solution *B* was included. Longevity was about equal in both; however, where technical sucrose (experiment 8) was used, approximately three times as many eggs were deposited. Technical sucrose may contain certain impurities that are essential for egg production.

When technical sucrose was placed in battery-jar cages in a dry condition (experiment 5) longevity was greatly reduced and fecundity was greatly increased. A total of 30 eggs per female was obtained, which was more than from any other experiment in the series. When placed in screen cages in trees in dry condition (experiment 7) practically all flies died before reaching egg-laying maturity. In this instance mortality was probably due to lack of moisture. Under identical conditions, except that a 10 per cent sucrose solution was used instead of dry sucrose (experiment 6), the longevity factor was approximately equal to that of battery-jar cages in the laboratory (experiment 1), while the egg production was increased three times.

Honey, in comparison to sucrose, in battery-jar cages (experiments 1 and 22) materially reduced longevity though it more than doubled egg production. In comparing honey with sucrose in quartz-glass cages (experiments 2 and 23) negligible differences existed in longevity, while slightly more eggs were deposited in the honey tests. Honey-yeast compared with sucrose-yeast (experiments 3 and 24) showed a negligible difference in longevity, but in the former egg production was increased nearly four times. Honey in screen cages in trees in comparison to sucrose under identical conditions (experiments 6 and 26) showed slightly reduced longevity, though the fecundity was nearly doubled.

When honey was fed in quartz-glass cages, in comparison to battery-jar cages (experiments 22 and 23), longevity was slightly increased though approximately half as many eggs were deposited.

Under screen cage conditions in the laboratory, as compared with screen cages in trees (experiments 25 and 26) with honey for food, the flies lived considerably longer and deposited four times as many eggs under the latter conditions. Screen cages in trees, both with honey and sucrose (experiments 6 and 26), as compared with battery-jar cages in the laboratory (experiments 1 and 22), appeared to produce conditions more conducive to egg production. These facts suggest a relation of sunlight or certain of its component rays to fecundity of the flies.

Several sugars were employed singly with mineral nutrient solution *B,* which included nitrogen, and also combinations of these sugars without minerals though with nitrogen as urea. Levulose plus solution *B* in comparison with sucrose plus *B* (experiments 14 and 18) reduced lon-

gevity materially, though egg production was more than doubled. Flies fed dextrose plus solution *B* lived only approximately half as long as those fed sucrose plus solution *B* (experiments 14 and 19); however, negligible differences existed in the numbers of eggs deposited. Dextrin plus solution *B* (experiment 20) was apparently of little value as food, since flies died nearly as rapidly as in the control tests of water and of solution *B* (experiments 27 and 28). In the combination of sucrose, levulose, dextrose, dextrin, and urea (experiment 17) longevity was slightly increased and fecundity was unaffected in comparison to levulose plus solution *B* (experiment 18), while in comparison with sucrose plus urea (experiment 15) the longevity factor was practically equivalent, though fecundity was nearly doubled. However, in the same combination minus sucrose (experiment 21) all flies died before reaching egg-laying maturity.

These tests indicate that sucrose, levulose, dextrose, or honey is essential for longevity and fecundity; that both honey and levulose reduced longevity slightly in comparison with sucrose, although fecundity was increased; that dextrose reduced longevity in comparison with sucrose without affecting fecundity; and that flies cannot survive on dextrin alone.

*Results with Minerals in Foods.*—Mineral nutrient solutions *A* and *B* are regular media employed in plant-nutrition research. The only difference between these solutions is the omission of zinc and copper in solution *B*. Experiments were planned in which each mineral would receive consideration; however it was not possible to carry these out. Sucrose plus solution *A*, when compared with sucrose plus solution *B* (experiments 13 and 14), showed a slight increase in longevity and doubling of egg production. Solution *A* added to sucrose-glycocoll (experiments 10 and 11) increased longevity slightly and doubled egg production. Solution *B* added to sucrose-glycocoll (experiments 10 and 12) slightly increased longevity and decreased fecundity. The data indicate somewhat increased longevity and fecundity as the result of zinc and copper in the diet.

*Results with Foods of Varying pH.*—The hydrogen-ion concentration of the various media ranged from pH 3.8 to pH 9.5. However these studies fail to show that any relation exists between longevity or fecundity and the pH of media throughout the range tested.

*Male Reproductive System.*—The male reproductive system is diagrammatically shown in figure 51. The internal portion of this system occupies most of the space in the posterior abdominal cavity. The testes are relatively large, yellowish, kidney-shaped organs surrounded by tracheae that appear to anastamose interiorly among the testicular folli-

cles. The testes unite directly with the seminal vesicles at their inner anterior ends, where a constriction occurs. The seminal vesicles are somewhat club-shaped enlargements of the vasa deferentia. One fairly large group of accessory glands, consisting of convoluted tubes, occurs at the

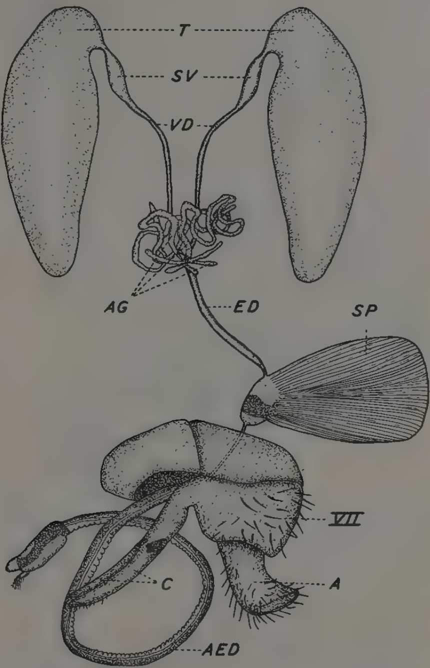



Fig. 51. Reproductive system and anus of male *Rhagoletis completa:* A, anus; AED, aedeagus; AG, accessory glands; C, claspers; ED, ejaculatory duct; SP, seminal pump; SV, seminal vesicles; T, testes; VD, vasa deferentia; VII, seventh abdominal segment.

union of the vasa deferentia and the ejaculatory duct. The seminal pump is a large, flattened, somewhat fan-shaped organ, consisting primarily of the chitinous ejaculatory apodeme, surrounded by muscular tissue. The ejaculatory duct passes through the proximal end of the seminal pump shortly before it protrudes from the body, where it becomes chitinized to form the aedeagus.

The external genital organs consist of the claspers and aedeagus. The claspers are a pair of prominent chitinous appendages on the venter of

the seventh abdominal segment. They serve to hold the ovipositor in place during copulation. The aedeagus is a long, coiled, chitinized organ

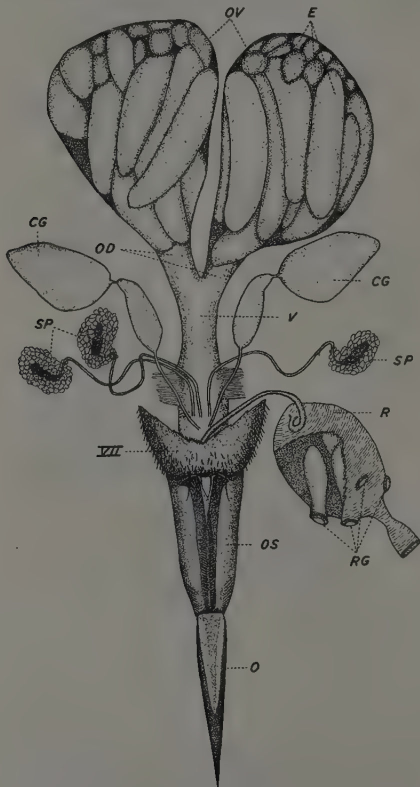



Fig. 52. Reproductive system and portion of of hind intestine of female *Rhagoletis completa:* CG, colleterial glands; E, ova in various stages of development; O, ovipositor; OS, ovipositor sheath; OD, oviducts; OV, ovaries; R, rectum; RG, rectal glands; SP, spermathecae; V, vagina; VII, seventh abdominal segment. (The arrangement of ova in the tubes is shown in fig. 53.)

attached to a supporting framework within the sixth and seventh abdominal segments. At the distal end it bears a small brush of stiff hairs.

*Female Reproductive System.*—The female reproductive system is diagrammatically shown in figure 52. The internal portion of this sys-

tem consists of two pyriform ovaries connected by short oviducts to the vagina, which leads into the ovipositor. There are three spermathecae and a pair of colleterial or accessory glands which lead into the distal portion of the vagina through ducts. Each ovary consists of approximately twenty-four egg tubes which are maintained in one unit by connective tissue and many branching tracheae. Each egg tube may contain a series of eggs in various stages of development from the germarium in the very anterior end where no differentiation is evident, to a mature egg in the posterior end (fig. 53); usually four or five eggs in various

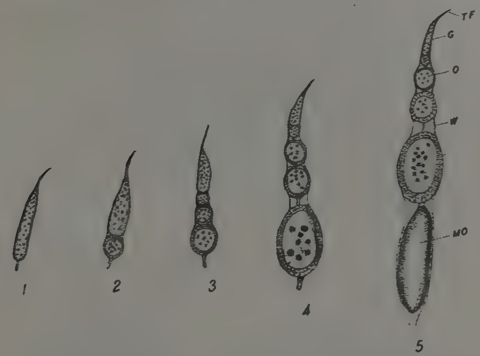



Fig. 53. Development of oöcytes in ovarian tubes with respect to number of days after female emerged from soil: 1, two days; 2, six days; 3, ten days; 4, fourteen days; 5, eighteen days. G, Germarium; MO, mature ovum; O, oöcyte; TF, terminal filament; W, wall of ovarian tube.

stages of development are present in each egg tube. When mature the eggs pass down the oviduct into the vagina, where fertilization takes place.

The external genital system is very simple, and consists of the ovipositor and sheath. The ovipositor is a slender sharp-pointed organ of chitinous structure with an opening on the ventral surface. It is attached to the last (VII) abdominal segment by the membranous sheath. This structure bears many short, triangular, chitinous projections on its surface. The ovipositor is usually telescoped into the sheath, which is in turn telescoped into the last abdominal segment.

*Copulation.*—Under laboratory cage conditions the time intervening normally between emergence of females and first coition with the male is from 7 to 14 days. An experiment was conducted to obtain information relative to the age of males and females at the time of copulation. Twenty-five males and a like number of females were used in each test.

In cage A newly emerged males were confined with 9-day-old virgin females. Six days later two pairs were observed copulating and fertile eggs were deposited 2 days after copulation. However, copulation was more common after 10 days had elapsed. This demonstrates that males are capâble of the act at least within 6 days after emergence.

In cage B, newly emerged females were confined with 9-day-old unmated males. The following day a male finally succeeded in copulating with one of the females, after a struggle on the part of the female to prevent the act. Each succeeding day for 4 days after the experiment began, females were observed struggling unsuccessfuly to prevent copulation. However, after the females were 4 days old, they did not seem to resist so vigorously. Fertile eggs were deposited within 12 days after the experiment began.

Under laboratory conditions, copulation has been recorded at every hour of daylight at temperatures from 60° to 103° F. In the field also it may take place at almost any time of the day under varying conditions. Normally it is more commonly observed in the late afternoon, since it usually follows oviposition. In midseason in a grove with a large fly population, after about 4 p.m., many of the walnuts will have one or two males perched upon them. It is very interesting to observe how a male maneuvers in an effort to keep other males off while waiting for a female to alight on the nut. When he first takes up his position on the nut in the afternoon he is usually not very active until after one or two combats, following which he patrols the nut very effectively. He walks spryly about, with wings extended slightly upward and outward, moving them by sudden jerks, much after the fashion of a strutting peacock.

When another male alights on the nut, the original occupant extends his wings high over the body and approaches his opponent cautiously, finally darting into him bodily in an effort to dislodge him; or they approach each other and fight by standing up on their hind legs (venter to venter) and using their fore and middle legs with which to strike (fig. 54 *B*). In many instances they lose their balance and both fall off the nut, in which case the original occupant usually returns immediately to resume his vigil. If they do not fall off, the battle sometimes lasts several minutes, until one either is dislodged or is cowered and flies away. On several occasions as many as five males have been observed in one group, fighting one another while standing on their hind legs. Often coccinellid beetles appearing on the nut in search of aphids are attacked and driven away by the male fly.

When a female alights on the nut, the male behaves very differently. He apparently becomes very excited, as is evidenced by his actions. The object of her presence there is to feed or oviposit, and she usually takes

up her normal actions pertaining thereto without any apparent notice of the male's presence. The male approaches the female very cautiously, moving quickly sideways and apparently directing his path of approach so as to overtake the female by surprise from the rear. In many instances the female has been observed to detect the male's presence, turn and dart into him bodily, and fly away after the collision. In such cases

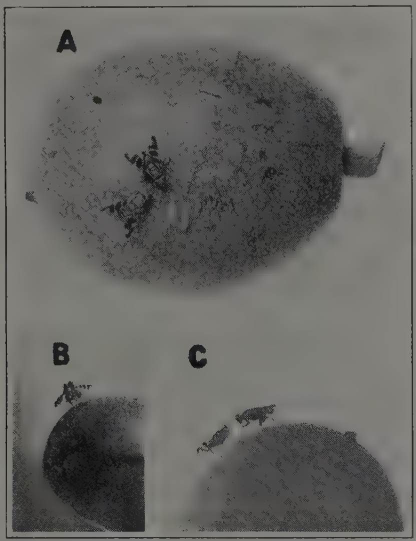



Fig. 54. Characteristic habits of adults of *Rhagoletis completa: A* and *C,* male and female in characteristic position on a walnut during oviposition; *B,* males fighting for supremacy of the walnut.

the female is probably either too young for copulation, is anxious to deposit a batch of eggs, or has recently been fertilized. However, the female is usually ready for copulation immediately after depositing eggs. During oviposition a male usually stands close by (fig. 54 *A–C*) or moves about, at no time getting very far away from the female. As soon as the ovipositor is withdrawn from the walnut husk, the male mounts her and generally no effort is made to resist him. She extends her wings laterally and raises the end of her abdomen, with ovipositor protruding, to aid in the union. With the hind pair of legs the male guides the tip of her abdomen so that the extended ovipositor comes to rest in the claspers of his genital organs. The intromittent organ or aedeagus is quickly

inserted and copulation ensues, usually occupying a period varying from 2 to 15 minutes. On several occasions a male has been observed to mount a female while she was in the act of oviposition and withdraw her ovipositor by force to enter into coition.

In virgin-female studies an experiment was conducted with respect to egg development in which unmated females were confined with males. Twenty-five females, 30 days old, were placed in a cage with as many males. The following afternoon, and each succeeding afternoon for several days, a few pairs were observed copulating. In each instance they remained in coition for periods of 30 to 40 minutes, which is much longer than in normal cases.

It is not definitely known how often copulation is necessary. In many observed cases, both in the field and in the laboratory, the females submitted to the act passively immediately after oviposition. This evidence, and the very promiscuous tendencies of the males indicate that it is of frequent necessity with females that are freely ovipositing, and probably takes place after depositing each batch of eggs. Under laboratory conditions in oviposition studies where one or two females were caged with males and with susceptible host material, copulation occurred very frequently, sometimes daily, over periods of time. However, only a few eggs, often none, were deposited during the lifetime of females even when they remained alive for 30 days or longer.

*Preoviposition Period.*—The development of eggs in the reproductive system, with reference to time elapsed after emergence, received considerable attention. This study had a very important bearing on the control of the fly. Flies of known age were dissected at two-day intervals in the early part of the study to obtain information pertaining to the rate of egg development.

The method of dissection that proved satisfactory and was used throughout was as follows: Regular embedding paraffin was melted and a layer approximately ¼ inch thick was allowed to harden in the deep-type Syracuse watch glass. The flies were killed with ether or cyanide and placed on their backs in rows on the hard paraffin. With a heated needle they were attached to the paraffin so as to cause them to remain stationary for dissection. Water was placed in the watch crystal to such a depth that the flies were completely submerged. All dissecting was performed with the aid of a wide-field binocular microscope. A dissecting needle, ground and honed to a cutting edge on the point, served to incise the abdomen and, with the aid of sharp-pointed forceps, to remove the digestive system and certain muscles and tracheae. The ovaries, which lie near the dorsal surface, are readily observed when the digestive system is removed.

For general studies a few egg tubes may be teased out with cactus-spine needles, and studied *in situ* under higher magnification of the binocular. However, for quick detailed studies, it is more satisfactory to remove an ovary and place it in some temporary mounting medium on a microscope slide. The egg tubes can be teased out of the mass under the binocular before the cover slip is placed over the preparation. The mount is then studied under the desired magnification with the compound microscope. When time was not available for immediate detailed study the ovaries were removed and preserved in 5 per cent chloral hydrate for subsequent examination.

As the study progressed, it became evident that four-day intervals were often enough to make dissections, since a period of this duration was required under prevailing conditions to permit marked changes in the developing eggs (fig. 53). This is in agreement with Illingworth[23] in his studies on *Rhagoletis pomonella*.

These dissections of the females showed that the time required under laboratory conditions for the first eggs to be completely developed in the reproductive system was from 10 to 20 days. There was considerable variation in degree of development among individuals of the same age. However, the average for all groups at the seasonal peak of field emergence was 18 days. The higher temperatures with certain limitations increase the rate of egg development, and lower temperatures have a retarding effect.

It is recognized that the conditions of confinement of the flies that were dissected may affect the normal development of the eggs. Back and Pemberton[2] report a relation between kind of food and rate of development of eggs of the melon fly of *Bactrocera cucurbitae* (Coq.). It is entirely possible that lack of certain rays of the solar spectrum together with lack of certain nutritive elements may have affected the flies adversely, resulting in reducing the rate of egg development. An effort was made to compare egg development in the inverted battery-jar cage generally used with that occurring under natural conditions and with that in the small screen cage and in the large cheesecloth cage over walnut trees.

Accordingly an experiment was set up, on the assumption that a 16-mesh wire screen cage over a small bearing Eureka walnut tree would afford natural conditions for the flies. Newly emerged flies were placed in the four different types of cages at the same time. Samples of 10 females from each cage were dissected at 4-day intervals, and the average degree of egg development compared. The first 16 days of the experiment indicated that the large screen cage had not afforded satisfactory environment for the flies, and that the small type of screen cage

was less favorable still. The experiment was duplicated in 1930, with similar results. The average period required for the first eggs to reach maturity in the different types of cages was as follows:

| | | |
|---|---|---|
| Battery jar, artificial food | | 18 days |
| Large cheesecloth cage over walnut tree, natural food | | 18 days |
| Large screen cage over walnut tree, natural food | (estimated) | 24 days |
| Small screen cage, artificial food | (estimated) | 30 days |

The egg development of virgin females was studied to a limited extent. Experiments conducted throughout one season, in which 175 virgin females were used, showed that the rate of egg development was approximately the same as that for females confined with males. This conclusion is based on dissection and oviposition data.

In these studies relative to length of preoviposition period, a total of over 1,000 flies were used, of which approximately 500 were dissected to observe egg development. It should be pointed out that many females never deposited any eggs at all, even though apparently the first eggs were fully developed within 12 to 14 days after emergence from the soil.

*Oviposition.*—The number of eggs produced by a female is a matter of conjecture. Considering the fact that each ovary consists of 24 egg tubes and each egg tube usually contains 4 or 5 developing eggs, the reproductive potential of the species appears to be relatively high. Dissections of females that were known to have deposited over 60 eggs showed no indication of degeneration of the germaria. It seems logical to assume that a female is capable of depositing eggs under favorable conditions as long as she is in good physical condition. Therefore a reliable basis on which to calculate the number of eggs a female may deposit does not exist. It seems probable, however, that under optimum conditions in the field, females may deposit from 200 to 400 eggs.

Experiments were conducted in the field laboratory in 1929 and 1930, in an attempt to obtain detailed data regarding the more important features of oviposition. Series I in 1929 consisted of 12 battery-jar cages in which 2 females and 3 males were confined. Series II in that year consisted of 12 wire-screen cages in which 2 females and 3 males were confined. In 1930, series I and II were battery-jar cages and wire-screen cages, respectively, as before, though 1 female and 2 males were used in series I. In each season all tests were started on the same day, with newly emerged flies. The standard food throughout was lump cane sugar. Moisture was supplied only in the battery-jar cages. Both food and moisture were supplied in the manner previously described. Susceptible host material was supplied by placing a bottle of water containing a twig with two walnuts in each cage. The method of handling

walnuts maintained them in a susceptible condition for a period of 10 to 14 days. However, they were changed weekly in order to supply as favorable condition for oviposition as possible. For brevity, only the data obtained from the 1930 oviposition studies are presented in table 13.

It is evident that battery-jar cages afforded more favorable conditions for oviposition than wire-screen cages. However, the results secured in battery-jar cages were unsatisfactory. The females were very erratic in oviposition, and some did not oviposit at all, though the average length of life apparently aproached the normal. In battery-jar cages in 1930 the average number of eggs per female was 18.7, while in screen cages it was 1.3. The greatest number of eggs per female was 84 in 7 cavities (cage 14, series I, 1930) and the average length of time between ovipositions was 3 days. In one instance eggs were deposited by flies that were 62 days old, and in a few instances by flies around 50 days old.

Throughout these experiments, particularly in the battery-jar cages, the general behavior of the flies appeared to be normal in all respects except in oviposition. They copulated frequently, which fact led to the suspicion that the females might be voiding their eggs in the cages. Close examination of the interior of the cages at frequent intervals failed to disclose a single egg lying about. On many occasions when the host material was removed from the oviposition record cages it was placed in a stock cage containing many ovipositing females, to determine its susceptibility. In all such tests eggs were deposited in the husk tissue.

The total evidence at hand indicates that under no condition of laboratory confinement employed in this whole biological study of *Rhagoletis completa* have the flies responded normally with respect to oviposition factors. For instance, under natural field conditions the average number of eggs per cavity was 14.9, based on a total of 665 cavities. Under laboratory conditions the average number of eggs from 679 cavities was 8.2. Furthermore only a few instances have been observed under natural conditions where a female made a cavity and failed to deposit eggs in it. This is a fairly common occurrence in the laboratory. In the field laboratory, where large stocks of flies have been kept in battery-jar cages each season for varying periods of time, the average number of cavities per enclosed female has been relatively low.

If the data obtained from oviposition experiments were considered representative, the reproductive potential would be very low. It is believed, however, that the conditions under which the experiments were conducted did not approach the optimum and thereby reduced the fecundity of the females.

# TABLE 13
## Laboratory Studies on Oviposition in 1930

| Cage No. | Age (in days) of ♀ when eggs were deposited | | | | | | | | | | | | | | | | | | | | | | | | | Average age of females at death, in days | Cavities | Eggs |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | 10 | 12 | 14 | 16 | 18 | 20 | 22 | 24 | 26 | 28 | 30 | 32 | 34 | 36 | 38 | 40 | 42 | 44 | 46 | 48 | 50 | 52 | 54 | 56 | 62 | | | |
| | Number of eggs per cavity | | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Series I, 1 ♀ + 2 ♂ per battery-jar cage; set up August 8 | | | | | | | | | | | | | | | | | | | | | | | | | | | | |
| 1 | .... | .... | .... | 7 | .... | 12 | 0 | .... | .... | 9 | 4 | .... | .... | .... | 7 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 53 | 6 | 39 |
| 2 | 0 | 4 | .... | .... | .... | .... | .... | .... | 11 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 49 | 3 | 15 |
| 3 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 16 | 0 | 0 |
| 4 | .... | .... | 3 | .... | 13 | 7 | .... | .... | 14 | .... | .... | .... | .... | .... | .... | .... | 10 | .... | .... | 9 | .... | .... | 14 | .... | .... | 61 | 7 | 70 |
| 5 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 54 | 0 | 0 |
| 6 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 39 | 0 | 0 |
| 7 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 11 | 0 | 0 |
| 8 | 5 | 0 | .... | .... | 14 | .... | .... | .... | .... | .... | 12 | 7 | .... | .... | .... | 6 | .... | .... | 10 | 0 | .... | .... | .... | .... | .... | 60 | 8 | 54 |
| 9 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 51 | 0 | 0 |
| 10 | .... | .... | .... | .... | .... | .... | .... | .... | .... | 6 | 6 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 33 | 2 | 12 |
| 11 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 19 | 0 | 0 |
| 12 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 14 | .... | .... | .... | 8 | .... | .... | .... | .... | .... | .... | .... | .... | 45 | 2 | 22 |
| 13 | .... | .... | 12 | .... | .... | .... | 3 | .... | 9 | 1 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 58 | 4 | 25 |
| 14 | .... | 14 | .... | .... | .... | 12 | .... | 7 | 13 | .... | .... | 10 | .... | 17 | .... | 11 | .... | .... | .... | .... | .... | .... | .... | .... | .... | 55 | 7 | 84 |
| 15 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 13 | 0 | 0 |
| 16 | .... | 3 | 0 | 8 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 41 | 3 | 11 |
| 17 | .... | .... | .... | .... | 9 | .... | 7 | .... | .... | 14 | .... | .... | .... | .... | .... | 2 | 6 | .... | 6 | .... | 11 | 1 | .... | 3 | 8 | 67 | 10 | 67 |
| 18 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 55 | 0 | 0 |
| 19 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 34 | 0 | 0 |
| 20 | .... | .... | .... | .... | 13 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 51 | 1 | 13 |
| 21 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 3 | .... | .... | 7 | .... | .... | .... | 57 | 2 | 10 |
| 22 | .... | .... | 6 | .... | .... | .... | .... | .... | .... | 14 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 44 | 2 | 20 |
| 23 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 14 | .... | .... | .... | .... | .... | .... | .... | .... | .... | 52 | 1 | 14 |
| 24 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 6 | 0 | 0 |
| 25 | .... | .... | .... | .... | .... | .... | 9 | .... | 2 | .... | .... | .... | .... | .... | .... | .... | 1 | .... | .... | .... | .... | .... | .... | .... | .... | 49 | 3 | 12 |
| Average | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 43 | 2.4 | 18.7 |

TABLE 13—(*Concluded*)

| Cage No. | Age (in days) of ♀ when eggs were deposited | | | | | | | | | | | | | | | | | | | | | | | | | Average age of females at death, in days | Cavities | Eggs |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | 10 | 12 | 14 | 16 | 18 | 20 | 22 | 24 | 26 | 28 | 30 | 32 | 34 | 36 | 38 | 40 | 42 | 44 | 46 | 48 | 50 | 52 | 54 | 56 | 62 | | | |
| | Number of eggs per cavity | | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Series II, 2 ♀ + 3 ♂ per screen cage; set up August 20 | | | | | | | | | | | | | | | | | | | | | | | | | | | | |
| 1 | .... | .... | .... | .... | .... | 3 | .... | .... | .... | .... | .... | .... | .... | .... | 10 | .... | .... | 2 | .... | .... | .... | .... | .... | .... | .... | 46 | 3 | 15 |
| 2 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 31 | 0 | 0 |
| 3 | .... | .... | .... | .... | .... | .... | .... | .... | 6 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 56 | 1 | 6 |
| 4 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 4 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 57 | 1 | 4 |
| 5 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 49 | 0 | 0 |
| 6 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 16 | 0 | 0 |
| 7 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 42 | 0 | 0 |
| 8 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 21 | 0 | 0 |
| 9 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 34 | 0 | 0 |
| 10 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 7 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 45 | 1 | 7 |
| 11 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 32 | 0 | 0 |
| 12 | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 38 | 0 | 0 |
| Average per ♀ | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 39 | 0.3 | 1.[illegible] |

Limited observational data indicate that oviposition takes place only during daylight; it may take place at any time during the day when temperature, humidity, and light conditions are favorable. Based on extensive records, oviposition takes place at temperatures from 65° to 103° F. It has never been observed at a relative humidity lower than 60 per cent. Under natural conditions females are usually ovipositing most actively in the late afternoon from about five o'clock until darkness. Exceptions to this occur on dull, overcast days, which are not common during the season of greatest oviposition activity. A relation thus appears to exist between relative humidity and time of oviposition; however, light intensity may also be a regulating factor, judging from the conditions existing when oviposition has been recorded. Under battery-jar cage conditions in the shaded screen laboratory, where the light does not reach normal intensity and the relative humidity is usually 90 per cent or more, oviposition is as commonly observed at midday as at any other time.

When ready to oviposit the female tests the surface of the walnut, apparently to find a suitable place. In doing so she usually goes through a very characteristic set of maneuvers. With proboscis extended she walks about making frequent contacts with this organ. She will often stop and bring the venter of her body to rest on the spot momentarily; sometimes with feet stationary the whole body will be moved quickly up and down, or from one side to the other, or a combination of both movements. When an area is chosen she turns around in a circle several times, often reversing direction, then raises the body well upward and with arched abdomen attempts to force the ovipositor into the tissue at an angle of about 45 degrees. The spot selected frequently proves to be too hard; however, she tests it a few times with the ovipositor before searching for another spot. In the field laboratory one female made four attempts to insert her ovipositor in the same small area of husk without success; another female immediately made several attempts to utilize the same area in vain; a third female followed and with considerable effort finally penetrated the tissue and deposited a batch of eggs. These observations may indicate a variation in muscular strength among individuals.

When a favorable spot is located, the ovipositor is forced into the tissue (fig. 54 *C*) to a depth of about 2 mm and the whole body is moved around in a semicircle, or, often, in a complete circle. This procedure lacerates the tissue below the surface by virtue of the angle assumed by the ovipositor and thereby prepares the cavity for the eggs. The eggs are deposited one at a time. The ovipositor is withdrawn slightly and then forced quickly downward in the nature of a jab. Coincident with

this, the abdomen appears to be contracted to force the egg downward and out the opening of the ovipositor into the cavity. The egg can be detected as it passes the semitransparent portion of the sheath. After each egg is deposited, the ovipositor is withdrawn slightly and usually the body is rotated somewhat to place the next egg in position within the cavity. The eggs are placed one beside another on end at approximately right angles to the surface. The time required for depositing a batch of eggs varies from 3 to 10 minutes. During oviposition the retracted mouth parts pulsate rapidly and continuously.

When flies were confined in cages with walnuts, on several occasions two or three eggs were noted lying on the surface of the walnut near a cavity containing eggs. Apparently the female discharged these eggs directly on the surface. In this connection, with citrus fruits, the females have been observed to discharge their eggs on the surface after unsuccessful attempts to make a cavity (fig. 24, p. 390). In one instance, on peppers, a batch of 18 eggs was observed on the surface near a cavity containing 6 eggs (fig. 27, p. 394). One female is believed to have deposited all of these eggs, continuing to discharge them on the surface after the cavity had been filled.

Usually one female deposits all the eggs contained in a single cavity and does so with one insertion of the ovipositor. In a few exceptional cases out of the hundreds examined in detail, 30 to 40 eggs were present, probably representing two contingents; that is, in approximately half of them the embryo was well developed, while with the remaining ones, practically no development had occurred, which indicated recent deposition. It is not known whether these eggs are the products of one or two females. One female may have deposited both batches at different times; for under laboratory conditions the same individual has been observed to return to an exact spot several days after previous unsuccessful attempts to penetrate the husk tissue for oviposition.

Many observations were recorded of females attempting to oviposit in citrus fruits. With ripe Valencia oranges, for instance, the skin was fairly readily penetrated by the ovipositor, but the tissue below the surface was of such texture that the female could not lacerate it. It was interesting to note how the female would increase the angle of insertion of the ovipositor upon being unable to turn the body around in the original position. The acuteness was usually reduced until the ovipositor was inserted at right angles to the surface, then the female could turn around with ease but without accomplishing the desired purpose. One female worked diligently for half an hour attempting to produce a cavity, then went away and later returned to the same spot and excitedly worked as long again without success. In such cases, however, one egg

was usually placed in the hole made by the ovipositor and others voided on the surface in the immediate vicinity (fig. 24, p. 390). Eggs in such locations dried up before hatching.

The behavior of the flies when confined on walnuts of less susceptible varieties was studied, and data were obtained to supplement field observations. Placentia walnuts were placed in cages with ovipositing flies. In but few instances were the flies capable of penetrating the outer surface of the husk with the ovipositor. In the successful cases observed, penetration was accomplished only after repeated attempts and considerable effort. After the ovipositor had penetrated the outer husk, little difficulty was experienced in lacerating the tissue below the surface. In view of the observations on oranges, where one or more females attempted to use the same puncture for oviposition, it was desirable to know whether or not they would take advantage of artificial punctures on walnuts. Accordingly on several occasions one or more small needle punctures were made on Placentia walnuts in the vicinity of unsuccessful attempts at oviposition by females. They frequently returned to this area to attempt oviposition, but in no instance did they make use of one of the artificial punctures.

Brooks[8] reports that *Rhagoletis suavis* usually takes advantage of abrasions and decaying tissue in ovipositing in walnut husks in eastern United States. Since the biology and habits of *completa* are somewhat similar to those recorded for *suavis,* it was of interest to determine whether or not *completa* would have similar habits, under laboratory conditions. Many records of oviposition in the field showed that *completa* always placed the eggs in healthy tissue, even when natural or artificial abrasions were present for the female to select. Under laboratory conditions Eureka walnuts, with different kinds of abrasions freshly made, and also in various states of decomposition, were placed in cages with ovipositing flies. In many instances the males would take up their vigil beside one of the freshly made artificial punctures, apparently expecting a female to utilize it in oviposition. The female surveyed the abrasions in selecting a spot to insert her eggs, but in no instance did she choose other than healthy tissue. The experiments were repeated many times, with similar results. When walnuts were removed from the cages the abrasions and decaying tissue were carefully examined for the presence of eggs, with negative results.

In one instance males were liberated in a cage containing 25 unmated females, some of which had oviposited previously. A considerable amount of copulation took place and a few batches of eggs were deposited, none of which proved to be fertile. Because of the lateness in the season and the resulting unfavorable condition of host material, the

studies were discontinued before further information was obtained. However, it is expected that fertile eggs would be produced under such conditions. The few batches of infertile eggs may have been deposited by unmated females since the flies were not marked and distinction was not possible.

Signs on the surface of the walnut husk indicating the presence of an egg cavity are very characteristic, both in appearance and location. The actual puncture made by the ovipositor cannot usually be detected with

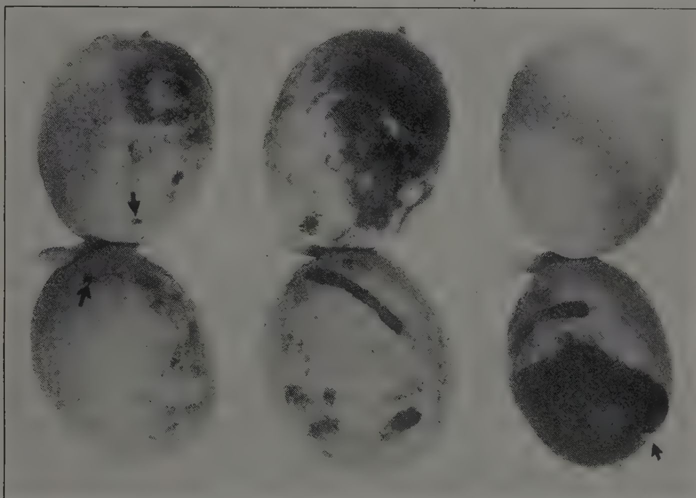


Fig. 55. General appearance of infested nuts in the field. Arrows at left indicate location of egg cavities. Arrow at right points to mature larva emerging from the husk. The so-called "tear stain" is shown on the lower walnuts in center and at right.

the unaided eye; but a droplet of colorless fluid exudes at the point of entry, which very rapidly dries. Flies of both sexes have been observed to imbibe this fluid. The lacerated tissue below the surface readily oxidizes and a resulting circular black spot several millimeters in diameter becomes evident externally (fig. 55). Another common sign a few days after oviposition is the presence of a black "tear stain" effect, resulting from small amounts of exuding husk sap which readily oxidizes after it has mixed with dew and trickled down the surface of the walnut (fig. 55). During the peak of seasonal activity of the fly, when prevailing temperatures are relatively high, the tissue broken down in making the

egg cavity becomes apparent as a dark spot within 1 hour. Later in the season the cavities are not usually detectable until after 12 to 24 hours have elapsed.

When the surface husk tissue over an egg cavity is removed, the eggs are plainly visible. Within several days after the female deposits a batch of eggs, the broken-down inner husk tissue resulting therefrom dries and shrinks, leaving a very perceptible cavity surrounding the eggs. The outline of the cavity is dark, as a result of oxidation, and affords a sharply contrasting background for the pearly-white eggs (fig. 7, p. 371).

For several years, during the peak of oviposition, data were collected regarding the number of egg cavities per walnut, with their respective positions on the surface, and also the number of eggs per cavity. A cavity located in the husk tissue in the anterior one-fourth of the nut was considered in the stem region; when located in the distal one-fourth, in the calyx region; and when located in the remaining central one-half, in the center. The position on the surface was further classified according to the exposure to light. If located so that the female was in the area of most light when ovipositing, often on the upper surface, it was considered outer; when in the area of least light, often on the lower surface, it was considered inner; and when neither of the above conditions prevailed it was considered neutral. The terms "outer" and "inner" were chosen rather than "upper" and "lower" because in many instances a cavity located on the lower surface was actually in position to receive most light, and vice versa. The observations were made on the Eureka variety. The data are presented in table 14.

The data for a 4-year period under field conditions show that 72 per cent of the egg cavities were located in the stem region; 24 per cent in the middle region; and 4 per cent in the calyx region. Under laboratory conditions, in 1931, the percentages are 36, 42, and 22 for the respective regions. Thus a wide difference existed between the relative locations of egg cavities under field and laboratory conditions. The husk-hardness data previously presented (pp. 380–388) show that in 1929 and 1930 the stem region was materially softer than the middle and calyx regions; but that in 1931 negligible differences existed between the hardness of the husk in various regions in Eureka walnuts. The field data for 1928, 1929, and 1930, and the laboratory data for 1931 to a lesser extent, indicate that the flies actually selected the softest regions of the husk in which to oviposit. However, this was not the case in the field in 1931. It is regrettable that detailed data regarding the location of egg cavities under laboratory conditions are not available for the first three seasons, in order to compare with field conditions. While it has been

TABLE 14

LOCATION OF EGG CAVITIES WITH RESPECT TO SURFACE, NUMBER OF EGGS PER CAVITY, AND NUMBER OF EGG CAVITIES PER WALNUT, UNDER FIELD AND LABORATORY CONDITIONS

| Year | Total cavities | Total walnuts | Location of egg cavity on surface of walnut | | | | | | Number of eggs per cavity | | | Cavities per walnut | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | Stem region | Center region | Calyx region | Outer | Inner | Neutral | Cavities | Total eggs | Eggs per cavity | 1 | 2 | 3 |
| | | | Cavities in per cent of total | | | Cavities in per cent of total | | | | | | Nuts in per cent of total | | |
| (Field) | | | | | | | | | | | | | | |
| 1928 | 320 | 300 | 70 | 14 | 16 | .... | .... | .... | 194 | 3,317 | 17 | 78 | 17 | 5 |
| 1929 | 348 | ...... | 60 | 36 | 4 | 12 | 32 | 56 | 116 | 1,707 | 15 | .... | .... | .... |
| 1930 | 435 | 216 | 74 | 23 | 3 | .... | .... | .... | 228 | 3,055 | 13 | 86 | 13 | 1 |
| 1931 | 861 | 645 | 76 | 22 | 2 | 13 | 45 | 42 | 127 | 1,816 | 14 | 72 | 23 | 5 |
| All years | 1,964 | 1,161 | 72 | 24 | 4 | 13 | 41 | 46 | 665 | 9,895 | 15 | 76 | 20 | 4 |
| (Laboratory) 1931 | 315 | ...... | 36 | 42 | 22 | .... | .... | .... | 679 | 5,589 | 8 | | | |
| Location of cavities with varying number of cavities per nut (field, 1931) | | | | | | | | | | | | | | |
| 1 cavity per nut | 462 | 462 | 73 | 25 | 2 | | | | | | | | | |
| 2 cavities per nut | 150 | 150 | 82 | 17 | 1 | | | | | | | | | |
| 3 cavities per nut | 33 | 11 | 70 | 27 | 3 | | | | | | | | | |

shown previously that husk hardness is probably the most important host factor governing oviposition, it cannot be considered the major factor with respect to location of egg cavities, in view of the contradictory field data for 1931. However, extensive field observations regarding oviposition show that undoubtedly husk hardness is a very important factor governing the spot where the eggs are deposited. In most instances observed the females vigorously attempted to insert the ovipositor at least several times in various regions of the husk before succeeding.

Data regarding the location of egg cavities with respect to outer, inner, or neutral positions show that only 13 per cent are placed in the area of most light. Limited husk-hardness data indicate insignificant differences in the hardness of these three locations. The females therefore actually do avoid the outer position for oviposition. Perhaps light intensity is an influencing factor.

The average number of eggs per cavity under field conditions was 14.9, in contrast to 8.2 in the laboratory. In the former instance 99.2 per cent of those cavities examined contained eggs, while in the latter they were present in 88.7 per cent of the cavities. Under field conditions the number of eggs per cavity varied from 4 to 40, while in the laboratory the variation was from 1 to 44 eggs.

Of the 1,161 infested walnuts examined over a 3-year period, 76 per cent exhibited 1 cavity per nut; 20 per cent had 2 cavities; and 4 per cent had 3 cavities. These data were obtained in heavily infested groves (90 per cent or over) where there was maximum opportunity for continued reinfestation. Under laboratory conditions in stock cages containing large numbers of ovipositing females, it was not uncommon to observe from 5 to 10 cavities per walnut, all made within a period of several days. The maximum number recorded per nut was 15. Females probably do not intentionally oviposit in walnuts that are already inhabited by a batch of larvae. Perhaps the repeated sampling on the surface of the nut with the proboscis prior to oviposition, as already described, is for the purpose of ascertaining whether or not larvae are present. In this connection it is of interest to note that after the larvae are about three-fourths mature a characteristic feeding noise is audible. In practically all instances where there were relatively large numbers of egg cavities per walnut under laboratory conditions the eggs were deposited before any larvae hatched.

In the 1930 field control studies, extensive observations were made on a total of 73 trees regarding the location of the infested nuts, that is, whether they occurred on the lower third, or the upper two-thirds of the tree. Of a total of 14,475 nuts examined on the lower portion of the tree,

12.2 per cent were infested. The middle and upper portions were necessarily grouped together, and of a total of 16,778 nuts examined, 17.5 per cent were infested. Therefore it appears that for oviposition the females show a slight preference for those nuts in the middle and upper portions of the tree.

## EGG

*Incubation.*—The incubation period for eggs was determined at the peak of oviposition under field conditions, and also under laboratory conditions for the same period. Under field conditions over 100 cavities were under observation, and complete and detailed information was obtained on 13 cavities, involving a total of 182 eggs. A mean temperature of 73° F, with a range of 52° to 104° F, prevailed for the first 5 days of the period of this study, during which time 85 per cent of the eggs hatched. The average time required for incubation was 120 hours, with a range of from 96 to 240 hours. Fragmentary data involving hundreds of eggs are in general accordance with the relatively few complete detailed records.

The laboratory studies, which yielded fairly accurate data, involved the use of 10 cavities, and a total of 123 eggs. The method used in incubating the eggs under artificial conditions was as follows: Newly deposited eggs were removed from the host tissue and placed on moist filter paper in petri dishes. They were kept darkened except when examined every 6 hours for evidence of hatching. The mean temperature prevailing for the same period of 5 days during which the field studies were made was 81° F, with a range of 75° to 89° F. The average time required for incubation was 72 hours, with a range of 48 to 120 hours. In this experiment many incomplete records likewise support the data obtained from the detailed studies.

*Mortality.*—In order to determine the mortality occurring naturally with eggs under field conditions, 112 egg cavities were carefully dissected under a binocular microscope. For these studies only nuts possessing one cavity were taken, in order to avoid the possibility of confusing the larvae of two batches of eggs. The eggshells and unhatched eggs were counted and recorded. The husk tissue was carefully dissected in order to determine the number of larvae present. If eggshells were present and none or only a very few larvae were inhabiting the husk, it was assumed that natural enemies had been active. When unhatched eggs were present together with eggshells, and the larvae of the batch were in the third instar, it was assumed that the remaining eggs would not hatch. It was not difficult to distinguish infertile eggs from fertile eggs in which the developing embryo had died. The infertile eggs usually

retained the pearly-white color until they were several weeks old, when they became semitransparent at both ends, their contents then being of watery appearance.

In these field mortality studies a total of 1,239 eggs was involved, of which 244, or 20 per cent, failed to hatch. Counts showed that natural enemies, probably the bug *Triphleps insidiosus* (Say) and the mite *Pediculoides ventricosus* New., or both, were responsible for 40 per cent of this total mortality, or a mortality of 12 per cent of all eggs.

Under artificial incubation conditions in the laboratory, using the method previously described, a total of 636 eggs was studied, of which 141, or 22 per cent, failed to hatch.

## LARVA

*Hatching.*—A few hours before the newly developed larva issues from the egg, movement can be observed. The dark-colored oral hooks and pharyngeal skeleton are very conspicuous through the now transparent chorion. To extricate itself the young larva ruptures the shell by actively scratching a small area on the interior a short distance below the point, and gradually works its body outward through this opening. When the anterior end of the body protrudes through this opening, the oral hooks are used to make firm contacts with the substratum to aid in pulling the body outward. The whole operation usually requires approximately one minute.

Under normal conditions the larva penetrates the host tissue immediately. When hatching under artificial conditions they move about aimlessly and in a few instances have been observed to tunnel between the layers of moistened filter paper. Without food they remain alive from 6 to 12 hours. Locomotion is accomplished through a coördination of the oral hooks, ventral fusiform areas, and the muscles of the individual body segments. The pointed oral hooks normally penetrate the substratum to insure a firm footing while the whole body is drawn forward, each segment being contracted. The ventral fusiform areas make firm contacts while the anterior end of the body is sent forward by the expanding body segments. Thus, essentially, locomotion consists of a series of waves of expansion and contraction of body segments.

*Feeding.*—The larvae show some tendency toward gregarious habits in feeding. This is not strictly the case, however. Many of the newly hatched larvae leave the egg cavity and enter the normal husk tissue through the same tunnel, but later make individual tunnels. These tunnels often come together and are otherwise usually within close proximity to one another in the particular area of husk tissue inhabited. The developing larvae have no tendency toward scavenger habits, always

showing a preference for healthy tissue for food. When the decaying tissue is all that is available in the area surrounding them, they consume it. This black food in the alimentary canal is plainly visible externally. After the larvae are mature, they are commonly found in a group in the area of broken-down tissue. Under these conditions it is doubtful if they are consuming any appreciable amount of this material for food, since they are usually of creamy-white color. They are probably congregated in this area because it offers less resistance to emergence from the nut for pupation (fig. 56).

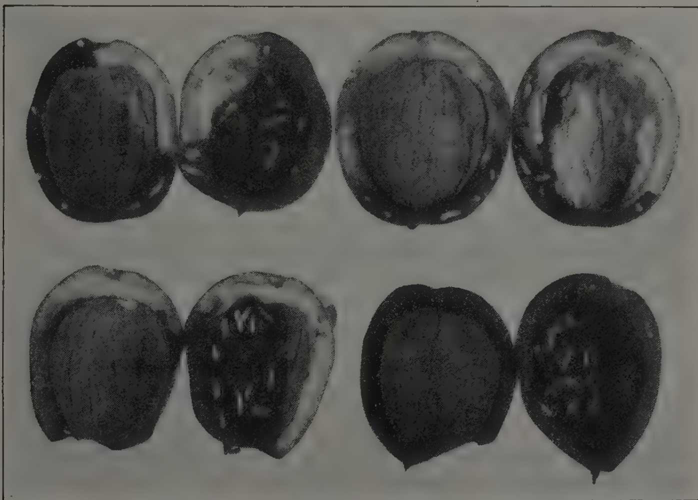


Fig. 56. Infested walnuts sectioned to show larvae *in situ* and characteristic destruction of husk with resultant stained shells.

The amount of food necessary to support a larva to maturity is relatively small. Detailed studies of random samples involving hundreds of walnuts were made in 1930, in which the number of practically mature larvae per nut was determined. The average was 12 larvae per walnut. In one instance, under cage conditions, 167 larvae were recorded from a single walnut. All larvae had attained the third instar, though they were somewhat smaller than normal. Externally the husk was entirely black. The inner husk tissue had been completely destroyed except for a few dry shreds. Other studies were conducted in 1930, to determine the area of stain on the shell of the nut as a measure of the amount of husk consumed. In all cases, except where the entire inner husk is consumed by the larvae, the amount of husk consumed is slightly more than the cor-

responding area of stain on the shell. These data are based on over 1,000 infested nuts and show that 65 per cent of the inhabiting larvae consumed one-half or less of the total inner husk while maturing, and only 14 per cent consumed the entire portion of this tissue.

*Development.*—Larval metamorphosis essentially consists of two molts or three instars. The methods employed in studying larval development were as follows: Eggs were permitted to hatch under artificial conditions. Sections of green Eureka husks ½ inch square were removed and placed (outer surface down) on moistened filter paper in large petri dishes. A small artificial tunnel was made in the inner tissue and one newly hatched larva placed in the opening. The petri dishes were kept darkened except when the contents were being examined. Over 500 first-instar larvae were used in these studies.

Preliminary observations established the approximate length of time between molts. Beginning 12 hours before the expected molt, samples consisting of 5 larvae were examined at 6-hour intervals to note the change in development. The section of host material was carefully dissected under the binocular microscope in an effort to locate the feeding larva. In many instances, particularly when dealing with the first instar, it was difficult to detect the larva before it was injured in the dissection. When uninjured they were transferred to other sections of fresh host tissue after noting the degree of development. After the first molt they were more readily detected and the mortality due to dissection was considerably reduced. After the second molt it became necessary to use larger sections of host tissue and to transfer the larvae every fourth day until they matured. When the larvae reached approximately three-fourths maturity, the mortality became relatively high again, primarily because of failure to remain in the host tissue. When 50 per cent of the larvae examined had transformed, the average elapsed time was considered an approximation of that required to complete the particular instar. It was necessary to establish an arbitrary criterion because of the variation in time required for larvae of the same batch of eggs to complete an instar. This indicates that they were not behaving normally under artificial conditions. In the field the development of larvae of one batch of eggs appears to be remarkably uniform.

In an attempt to approximate field conditions for purposes of comparison with rates of development under indoor-laboratory conditions, the petri dishes were kept in the standard hygrothermograph shelter in a walnut grove. Thus a record was obtained of the actual temperature conditions to which the larvae were subjected. The data obtained regarding larval development, both indoors and in the field, are summarized in table 15.

General field studies show that an appreciable number of larvae of the early portion of the brood may complete their development in from 18 to 20 days. This information was secured by tagging a large number of nuts in several groves, and also from observations on caged trees. Prevailing temperature conditions and a more succulent host tissue at this time are no doubt responsible for this shortened developmental period.

After the larvae have attained about three-fourths maturity a very characteristic feeding noise can be detected when in a quiet room, or by holding the infested nut near the ear in the field.

TABLE 15

DEVELOPMENT OF LARVAE UNDER ARTIFICIAL CONDITIONS IN THE INDOOR LABORATORY AND IN THE FIELD LABORATORY

| | Mean temperature | Range | Mean time for development | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | First instar | | Second instar | | Third instar | | Total development | |
| | *degrees F* | *degrees F* | *hours* | *days* | *hours* | *days* | *hours* | *days* | *hours* | *days* |
| Indoor laboratory | 82 | 67 to 90 | 127 | 5.3 | 199 | 8.3 | 343 | 14.3 | 669 | 27 9 |
| Field laboratory | 64 | 45 to 104 | 233 | 9.7 | 312 | 13.0 | 338 | 14.1 | 883 | 36.8 |

*Premature Mortality.*—Larval mortality during development was fairly accurately determined. At about the peak of larval maturity infested nuts were collected and studied in detail. Data were recorded only from those nuts containing larvae that were practically mature, none having left to pupate. Furthermore, unless the egg cavity or cavities could be definitely located and contents studied, the walnut was discarded. The difference between numbers of empty eggshells, less 12 per cent allowed for the work of natural enemies, and the mature larvae present, was considered to represent the mortality occurring during larval development. Data were obtained from 104 nuts. From 1,610 hatched eggs, 1,209 mature larvae were recorded. The mortality for the lot was 401 larvae, or 24.8 per cent. The average number of mature larvae per nut was 11.6; thus the average mortality was 3.9 larvae per nut.

*Emergence from Walnuts.*—When the larvae reach maturity they usually tend to congregate in the area of broken-down inner husk tissue, where they remain for a short while before emerging. In some instances this area includes the cavity that contained the eggs from which the

larvae originated. In such cases the puncture made by the female for oviposition is usually taken advantage of by the larvae in their exit. The size of the original opening is too small to detect with the unaided eye, but subsequent degeneration of the tissue often results in an opening from ½ to 1 millimeter in diameter. In any case it usually serves as a basis for the exit hole since it is fairly readily enlarged by rupturing the surrounding outer husk tissue. This tissue is now usually of a thin leathery texture. If an opening does not already exist, the larvae apparently are capable of making one with their oral hooks. In no instance has any evidence been found of larvae having emerged from a walnut through healthy tissue.

All larvae of one batch of eggs usually emerge through the same small opening. The oral hooks are placed through the opening to make contact on the outer surface. This serves to draw the first few body segments through the opening until the size of the hole prevents further ready penetration. The outward-projecting anterior portion of the body is then rotated in as wide an arc as possible, which serves to draw a portion of the plastic body segment through the hole at an angle. The emerging larva seems to force the body contents forward into the projecting anterior end as much as possible to aid in the exit. From 3 to 15 minutes is usually required for a larva to extricate itself completely from a nut under these conditions. When finally freed, it drops to the ground. All larvae within a single husk do not emerge one immediately following the other. In fact several days may elapse after the first larva emerges before all larvae have departed from the nut, particularly when only one exit hole is utilized.

In some instances when the uninjured portion of the husk of an infested walnut opens characteristically in ripening, the mature larvae leave through the split in the husk. This occurs only when the split in the uninjured husk is very close to injured tissue, which leaves a direct opening to the outside.

In the regular harvesting process the trees are systematically shaken and the infested nuts generally drop with husks adhering. The husk usually splits as a result of the impact with the ground, particularly if the nut contains mature larvae. Under these conditions the larvae emerge at about the same time and enter the soil immediately. It commonly happens that, because of the decaying husk tissue around the stem region, the walnut with husk attached pulls away from the stem at this point, the stem remaining on the tree. This results in a fairly large opening through which the larvae emerge if the husk is not ruptured in their immediate vicinity. Because of the possible bearing on control measures, an effort was made to determine the approximate percentage

of nuts from which the larvae had emerged at time of harvest for the Eureka variety of walnut. The data obtained are summarized in table 16.

The time of harvest in 1929 and in 1932 was earlier than that for 1930 and 1931. Furthermore it was considerably earlier in 1932 than in 1929. The data clearly show that the earlier the harvest the greater the number of walnuts that still contain larvae.

Occasionally the injured husk dries and hardens around one or more larvae, thereby trapping them. Since larvae are capable of pupating shortly after the third instar is reached, they usually enter the pupal stage under such conditions. In certain detailed studies involving an examination of every nut on a medium-sized infested tree, together with the larvae and pupae present in the nuts and in the soil, it was found that 0.14 per cent of the total pupae occurred beneath the dried husk, where the larvae had been trapped and had pupated.

TABLE 16

SEASONAL COMPARISON OF NUMBER OF INFESTED WALNUTS FROM WHICH LARVAL EMERGENCE HAD OCCURRED WHEN HARVEST BEGAN

| Year | Date | Total walnuts examined | Per cent of walnuts from which larvae had emerged |
|---|---|---|---|
| 1929 | October 4 | 1,124 | 43 |
| 1930 | October 15 | 2,408 | 77 |
| 1931 | October 10 | 1,818 | 87 |
| 1932 | September 25 | 2,790 | 25 |

Under natural conditions in the field, larvae have been observed to emerge from the nuts at all hours of the day. However, they were more commonly noted emerging in the cooler portions of the day. In several instances larvae were observed to drop onto the soil in direct sunlight during midday, where they were killed by the high temperature before they could enter the soil. This suggested the possibility of a relation between larval emergence and time of day with specific reference to temperature. An experiment was conducted in an attempt to obtain information relating to this matter. The soil beneath two medium-sized trees in a heavily infested grove was covered with canvas, in order to collect the larvae periodically as they dropped from the nuts. These records, together with prevailing air temperature and humidity, are presented in table 17.

The greater percentage of larvae emerged in the morning, between the hours of 5:30 and 8:00 o'clock, on both days. Practically the entire emergence on both days occurred when the temperature range was from

40° to 71° F, with the range of relative humidity from 95 to 40 per cent. Larvae did not emerge during the night, although the temperature and relative humidity apparently were not unfavorable. Throughout the greater part of both nights the temperature was not below 45 degrees nor the relative humidity below 80 per cent. It seems probable that temperature would bear more of a relation to time of larval emergence than would humidity; and, if true, the limits of favorable temperature are fairly narrow.

TABLE 17

Time of Day When Larvae Emerged from Walnuts; 1931

| Hour of day | Temperature, degrees F | Relative humidity, in per cent | Tree No. 1 | Tree No. 2 | Both trees | Per cent of total larvae per day |
|---|---|---|---|---|---|---|
| | | | Number of larvae | | | |
| September 21 | | | | | | |
| Before 5 a.m. | 46 | 87 | 0 | 0 | 0 | 0.0 |
| 5–6 a.m. | 47 | 80 | 195 | 270 | 465 | 33.4 |
| 6–7 a.m. | 58 | 56 | 284 | 289 | 573 | 41.2 |
| 7–8 a.m. | 71 | 40 | 160 | 113 | 273 | 19.6 |
| 8–9 a.m. | 80 | 35 | 26 | 14 | 40 | 2.9 |
| 9–10 a.m. | 82 | 28 | 10 | 7 | 17 | 1.2 |
| 10–11 a.m. | 87 | 24 | 3 | 7 | 10 | 0.7 |
| 11 a.m.–12 m. | 88 | 23 | 1 | 2 | 3 | 0.2 |
| 12 m.–1 p.m. | 90 | 22 | 1 | 6 | 7 | 0.5 |
| 1–3 p.m. | 85 | 26 | 2 | 2 | 4 | 0.3 |
| 3–5 p.m. | 77 | 38 | 0 | 0 | 0 | 0.0 |
| Total | ........ | ........ | 682 | 710 | 1,392 | 100.0 |
| September 22 | | | | | | |
| Before 5 a.m. | 41 | 95 | 0 | 0 | 0 | 0.0 |
| 5–6 a.m. | 40 | 94 | 85 | 55 | 140 | 7.7 |
| 6–7 a.m. | 49 | 69 | 295 | 310 | 605 | 33.4 |
| 7–8 a.m. | 58 | 50 | 400 | 290 | 690 | 38.1 |
| 8–9 a.m. | 66 | 42 | 136 | 238 | 374 | 20.7 |
| 9–10 a.m. | 74 | 36 | 0 | 0 | 0 | 0.0 |
| 10 a.m.–5 p.m. | 71 | 60 | 0 | 0 | 0 | 0.0 |
| Total | ........ | ........ | 916 | 893 | 1,809 | 100.0 |

*Tropic Responses of the Larva.*—Mature larvae show marked positive geotropism. This is an important factor in the preservation of the species. Under favorable conditions in light or darkness they disappear below the surface of the soil within 1 to 5 minutes after dropping thereon from their host. Their downward migration is not always perpendicular, for they appear to take advantage of the path of least resistance within

certain limits. Larvae in the second instar, and in the early portion of the third instar, do not exhibit such a marked geotropic response as do mature larvae.

Larvae in all stages appear to be positively thigmotropic, though to a limited degree. Perhaps it is in reality a geotropic response, for the conditions of the observations were not such that a fine distinction could be made. Whenever larvae were contained in petri dishes in which they were originally placed on moistened filter paper, approximately 25 per cent of them would find their way between and beneath the sections of paper within 24 hours. The behavior was similar in darkness and in daylight.

A simple method was employed to obtain information relative to the phototropic reactions of larvae. A wooden tray, 18 inches square and 2½ inches deep, was prepared as follows: One-half of the tray was lined with black cloth and also covered with the same material; the remaining half was lined with white cloth and left without a covering. With the tray orientated so that the black end was northward, lots consisting of 100 mature larvae were placed in the center of the tray within the confines of a small circle drawn with pencil. In this way one-half of the larvae were placed on the black cloth, just within the edge of the darkened end, the others being on white cloth and in daylight. Six hundred larvae were used in these tests. The number of larvae in each end of the tray was counted after they had dispersed from the center. An average of 81 per cent migrated to the darkened end, and the remaining 19 per cent moved about in daylight. After the limit of the darkened end had been reached, many of the larvae wandered aimlessly about, often returning to the daylight end. This test indicates that mature larvae are inclined toward negative phototropism.

A similar test was conducted in a regular photographic dark room, except that soil was placed in the bottom of the tray and a 50-watt electric light centrally located above the tray provided the only source of illumination. It was thought that if the larvae exhibited negative phototropism under these conditions they would enter the soil after migrating into the darkened end, and vice versa. Two hundred mature larvae were liberated in the center of the tray on the soil. There was very little migration in any direction except downward. After pupation, the soil was sifted in sections and 57 per cent of the pupae were taken in the soil of the dark end, while the remaining 43 per cent were in the light end. Under the conditions of this test, the positive geotropic response was probably so great that any significant tendencies toward phototropic responses were masked. However, burrowing into the soil may have been a negative phototropic response.

*Entering Soil.*—When the mature larvae come into contact with soil, they begin to burrow downward immediately, or as soon as a crack or crevice is found in which to obtain a firm hold with the mouth hooks. Penetration is effected in much the same manner as in migration on smooth surfaces. Occasionally a larva has been observed to enter the soil to a depth of about one-half its length, and when further penetration appeared difficult, to rotate the protruding posterior end, inscribing an arc in the same manner as with the anterior end when emerging from a small hole in a walnut husk.

TABLE 18

DEPTH TO WHICH LARVAE NORMALLY ENTER ORCHARD SOIL TO PUPATE

| Depth, in inches | Grove A Hanford fine sandy loam | | Grove B Yolo clay loam | |
|---|---|---|---|---|
| | Pupae* | Per cent of total | Pupae† | Per cent of total |
| 1 | 1,286 | 74.0 | 54 | 12.0 |
| 2 | 371 | 21.0 | 118 | 26.0 |
| 3 | 57 | 3.0 | 207 | 46.0 |
| 4 | 19 | 1.0 | 58 | 13.0 |
| 5 | 7 | 0.4 | 13 | 3.0 |
| 6 | 1 | 0.1 | 3 | 0.7 |
| 7 | 1 | 0.1 | 1 | 0.2 |
| 8 | 0 | 0.0 | 0 | 0.0 |
| 9 | 0 | 0.0 | 0 | 0.0 |
| Total | 1,742 | 100.0 | 454 | 100 0 |

* Eight plots of 1 sq. ft. each.
† Four plots of 1 sq. ft. each.

The loamy types of soil in the infested area under normal tillage at harvest time are readily penetrated by the larvae. These same soils may be packed by hand to such an extent that larvae are unable to penetrate them. Extensive sifting tests were made to determine the depth that larvae penetrate orchard soil under natural conditions to pupate. One square foot of soil in various locations was carefully excavated in layers 1 inch in thickness and sifted, and the number of pupae contained in each layer recorded. A total of 8 square feet of soil was sifted in grove A, and 4 square feet of soil in grove B. These data are presented in table 18.

In grove A the greater percentage of larvae did not penetrate the soil deeper than 1 inch; while in grove B the greatest percentage of larvae were found at the 3-inch level. Aside from difference in soil type, the surface soil in grove B was loose and very dry at the time of larval emergence from the walnuts. In both groves a few larvae reached the depth of 7 inches before pupating.

Within the infested area walnuts are found growing in five different types of soil: Hanford fine sandy loam, Hanford fine sand, Hanford sand, Yolo clay loam, and Chino clay adobe. It was desirable to study the effect of soil type on the larvae with respect to relative ease of penetration, depth of penetration, and mortality after penetration but before pupation. Samples of these types of soils were collected, sifted, and packed in a standard manner to a depth of 9 inches in battery jars. Sifting of the Chino clay adobe was not practical; samples were therefore taken from a bare area adjoining a walnut grove. An effort was made to maintain the samples practically comparable with respect to moisture present. All tests were set up in duplicate. One hundred and fifty mature larvae taken directly from host tissue were dropped onto the surface of the soil in each jar. The jars were arranged on a table under a large walnut tree and were not moved after the larvae were placed on the soil. A section of plate glass was used to cover the jars, to prevent other larvae from dropping into the jars and also to conserve the moisture present.

Within 2 hours after the experiment was set up, 95 per cent of the larvae had entered all types of soil except the Hanford sand. Here they moved around a great deal on the surface without being able to effect penetration. The sand shifted with them and the greater percentage of the larvae were unable to go downward. When this was observed, another jar of Hanford sand was set up and wetted until penetration by water had taken place to a depth of 6 inches. One hundred larvae were placed on the surface and within 1 hour all but three larvae had disappeared. Water causes the sand to cohere sufficiently for the larvae to make the firm contact necessary to force their bodies downward. The results of the penetration experiments are summarized in table 19.

Hanford sand was the only soil type that materially interfered with normal larval penetration. Furthermore in this soil mortality after pupation was very high. The explanation for this fact is lacking; perhaps weakening of the larvae by the several days of continuous activity in attempting to enter the sand partly accounts for it. The following experiment furnishes contributory evidence regarding this matter: Orchard soil was sifted and packed in a standard manner in a battery jar. In another jar the procedure was identical, except for further packing of the surface with the bottom of a bottle. This left the surface fairly hard. One hundred and seventy-five larvae were dropped on the soil surface in each jar. In the first jar they penetrated the soil within an hour; while in the second jar none was successful. However, in the second jar 9 per cent of the larvae had pupated on the surface within 24 hours, and 93 per cent within 68 hours. The larval mortality was 7

## TABLE 19

RELATION OF CERTAIN TYPES OF SOIL TO LARVAL PENETRATION

| Soil type | Test number | Mortality on surface, per cent | Pupated on surface, per cent | Per cent of total larvae that reached various depths in soil | | | | | | | Total pupae | | Mortality in soil before pupation, in per cent |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | 0–½ inch | ½–1 inch | 1–1½ inches | 1½–2 inches | 2–2½ inches | 2½–3 inches | 3–3½ inches | Number | Per cent | |
| Hanford fine sandy loam | 1 | 2 | 0.0 | 0.0 | 15.0 | 29.0 | 28.0 | 12.0 | 6.0 | 0.7 | 137 | 91 | 7.00 |
| | 1*a* | 2 | 1.0 | 3.0 | 12.0 | 28.0 | 37.0 | 0.0 | 0.0 | 0.0 | 123 | 82 | 15.00 |
| Hanford fine sand | 2 | 1 | 0.7 | 0.7 | 4.0 | 28.0 | 45.0 | 15.0 | 2.0 | 0.0 | 144 | 96 | 0.01 |
| | 2*a* | 0 | 0.0 | 1.0 | 2.0 | 19.0 | 41.0 | 19.0 | 9.0 | 1.0 | 137 | 91 | 0.10 |
| Hanford sand | 3 | 44 | 55.0* | 0.0 | 0.0 | 0.7 | 0.0 | 0.0 | 0 0 | 0.0 | 84 | 56 | 0.00 |
| | 3*a* | 38 | 59.0† | 0.0 | 1.0 | 0.0 | 0.0 | 0.0 | 1.0 | 0.0 | 93 | 62 | 0.00 |
| Yolo clay loam | 4 | 1 | 1.0 | 1.0 | 12.0 | 53.0 | 20.0 | 4.0 | 1.0 | 0.0 | 140 | 93 | 0.04 |
| | 4*a* | 0 | 0.0 | 0.7 | 13.0 | 26.0 | 55.0 | 2 0 | 0.7 | 0.0 | 146 | 97 | 0.03 |
| Chino clay adobe | 5 | 0 | 0.7 | 4.0 | 33.0 | 22.0 | 0.7 | 0.0 | 0.0 | 0.0 | 90‡ | 60 | 40.00 |
| | 5*a* | 2 | 1.0 | 14.0 | 28.0 | 12.0 | 0.7 | 0.0 | 0.0 | 0.0 | 84‡ | 56 | 42.00 |

* Eighty-two individuals dead, 1 alive.

† Perfectly formed puparia, though insects dead within.

‡ A few pupae were undoubtedly not accounted for, because of inability to pulverize all soil completely.

per cent. Practically all of the pupae were alive and apparently normal. In this test the larvae moved around on the surface attempting to enter, but less energy seemed to be required than when migrating on sand that was constantly shifting under them.

Chino clay adobe is a very heavy type of soil, and naturally offers great resistance to penetration. This fact may account for the relative shallowness of penetration and the increased mortality in comparison with the other three more favorable types of soil. In this adobe soil, the greater percentage of larvae penetrated to a depth of from 1½ to 2 inches.

*Laboratory Studies Regarding Effect of Temperature Upon the Mature Larva.*—Preliminary laboratory experiments were conducted to determine the effect of temperature upon the mature larva. For each individual experiment, 100 larvae were taken directly from walnut husks when the test was begun. Petri dishes 3½ inches in diameter and ½ inch deep, with several pieces of filter paper placed in the lower section, were used for containers. At the beginning of each test the filter paper was saturated with distilled water. In certain tests this moist condition was maintained throughout the experiment, while in others the paper was not moistened again during the period of the particular test. Temperature was maintained fairly constant though the equipment available did not permit entirely satisfactory control. A summary of the details of these experiments and the results obtained are presented in table 20.

At 30° F the exposure varied from 35 hours to 110 hours and in all instances mortality was materially increased over that in the controls. However, there was no consistent relation between length of exposure and mortality. In all instances after exposure to a temperature of 30° F, when removed and placed at the same temperature as the control (72° F), pupation was materially stimulated as compared with the control. However, a high percentage of the larvae died within the puparium shortly after it was formed.

Considering the total elapsed time, there was no significant difference in larval mortality between the tests at 36° and 42° F and the control at 72° F. At 50° F, after 150 hours' exposure, pupation was delayed somewhat, though mortality was approximately equivalent to that of the control. At this same temperature, after 800 hours' exposure, mortality was high shortly after pupation.

At 72° F (control) in the dry container, mortality was very high and consequently few larvae pupated. In the wet container no larvae pupated before 100 hours' exposure; after 300 hours' exposure there were 14 per cent live larvae, 12 per cent dead larvae, 62 per cent live pupae,

TABLE 20

EFFECT OF TEMPERATURE UPON MATURE LARVAE

| Experiment No.* | Moisture condition in container | Temperature | Exposure | Time at 72° F after removal from experimental temperature chamber: or length of exposure† | | | | | | | | | | | | Remarks |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | 24 hours | | | 48 hours | | | 100 hours | | | 150 hours | | | |
| | | | | Mortality, larvae | Pupated | | Mortality, larvae | Pupated | | Mortality, larvae | Pupated | | Mortality, larvae | Pupated | | |
| | | | | | Alive | Mortality | | Alive | Mortality | | Alive | Mortality | | Alive | Mortality | |
| | | *degrees F* | *hours* | *per cent* | *per cent* | *per cent* | *per cent* | *per cent* | *per cent* | *per cent* | *per cent* | *per cent* | *per cent* | *per cent* | *per cent* | |
| *A* | Moist‡ | 30 | 35 | 8 | 12 | 0 | 8 | 20 | 16 | 20 | 32 | 16 | 28 | 28 | 20 | Larvae frozen stiff after 24 hours' exposure |
| *A1* | Moist | | 45 | 40 | 8 | 4 | 44 | 12 | 8 | 52 | 4 | 16 | .... | .... | .... | |
| *A2* | Dry‖ | | 55 | 14 | 2 | 0 | 40 | 6 | 0 | 44 | 4 | 2 | 92 | 2 | 4 | |
| *A2a* | Moist | | 55 | 16 | 10 | 0 | 32 | 20 | 8 | 48 | 22 | 12 | 66 | 4 | 30 | |
| *A3* | Moist | | 72 | 0 | 8 | 0 | 24 | 32 | 8 | 36 | 32 | 12 | 48 | 20 | 32 | |
| *A4* | Moist | | 110 | 0 | 0 | 0 | 24 | 0 | 0 | 40 | 0 | 0 | 72 | 8 | 0 | |
| *B* | Moist | 36 | 110 | 2 | 2 | 0 | 4 | 14 | 0 | 4 | 20 | 0 | 6 | 34 | 0 | Larvae motionless at this temperature |
| *B1* | | | 135 | 0 | 0 | 0 | 2 | 0 | 0 | 4 | 10 | 0 | 16 | 30 | 0 | |
| *C* | Moist | 42 | 150 | 2 | 12 | 0 | 2 | 12 | 0 | 4 | 18 | 0 | 10 | 24 | 0 | Larvae move very slowly. Forty-two per cent pupated in chamber after 775 hours' exposure |
| *C1* | | | 800 | 6 | 50 | 0 | 10 | 54 | 20 | 18 | 42 | 26 | 18 | 38 | 44 | |
| *D* | Moist | 50 | † | 0 | 0 | 0 | 0 | 0 | 0 | 4 | 0 | 0 | 4 | 0 | 0 | Larvae move slowly. Six per cent dead and 24 per cent pupated in chamber after 550 hours' exposure |
| *D1* | | | 800 | 8 | 42 | 22 | .... | .... | .... | .... | .... | .... | 30 | 36 | 34 | |

* 100 larvae in each experiment.

† Heading "Length of Exposure" applies to experiments *D*, *E*, *E1*, *F*, *F1*, *G*, *G1*.

‡ "Moist" indicates that water was applied as required to maintain a fairly constant amount of moisture inside petri dish.

‖ "Dry" indicates that no water was applied to filter paper inside petri dishes after experiment was begun.

TABLE 20—(*Concluded*)

| Experiment No.* | Moisture condition in container | Temperature | Exposure | Time at 72° F after removal from experimental temperature chamber: or length of exposure† | | | | | | | | | | | | Remarks |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | 24 hours | | | 48 hours | | | 100 hours | | | 150 hours | | | |
| | | | | Mortality, larvae | Pupated | | Mortality, larvae | Pupated | | Mortality, larvae | Pupated | | Mortality, larvae | Pupated | | |
| | | | | | Alive | Mortality | | Alive | Mortality | | Alive | Mortality | | Alive | Mortality | |
| | | *degrees F* | *hours* | *per cent* | *per cent* | *per cent* | *per cent* | *per cent* | *per cent* | *per cent* | *per cent* | *per cent* | *per cent* | *per cent* | *per cent* | |
| Controls *E* | Dry‖ | 72 | † | 2 | 0 | 0 | 8 | 0 | 0 | 12 | 0 | 0 | 12 | 8 | 0 | Eighty-eight per cent dead larvae. Six per cent dead pupae, and 6 per cent live pupae after 250 hours |
| Controls *E*1 | Moist‡ | 72 | † | 4 | 0 | 0 | 4 | 0 | 0 | 4 | 18 | 0 | 4 | 24 | 0 | Twelve per cent dead larvae, 12 per cent dead pupae, and 62 per cent live pupae after 300 hours |
| *F* | Dry | 90 | † | 24 | 0 | 0 | 34 | 0 | 0 | 100 | 0 | 0 | ... | ... | ... | Larvae move actively and constantly |
| *F*1 | Moist | 90 | † | 0 | 0 | 0 | 0 | 0 | 0 | 10 | 4 | 0 | 30 | 18 | 0 | Larvae move actively and constantly |
| *G* | Dry | 100 | † | 14 | 0 | 0 | 92 | 0 | 0 | 100 | 0 | 0 | ... | ... | ... | Larvae move very actively and constantly |
| *G*1 | Moist | 100 | † | 2 | 0 | 0 | 40 | 0 | 0 | 100 | 0 | 0 | ... | ... | ... | Larvae move very actively and constantly |
| *H* | Moist | 115 | ½ | 8 | 0 | 0 | 12 | 2 | 0 | 20 | 10 | 0 | 52 | 16 | 0 | Larvae move very actively and constantly |
| *H*1 | Moist | 115 | ¾ | 100¶ | ... | ... | ... | ... | ... | ... | ... | ... | ... | ... | ... | Larvae move very actively and constantly |
| *I* | Moist | 125 | ½ | 84 | ... | ... | 92 | 0 | 0 | 94 | 0 | 0 | 100 | 0 | 0 | Larvae move very actively and constantly |
| *I*1 | Moist | 125 | ¾ | 100¶ | ... | ... | ... | ... | ... | ... | ... | ... | ... | ... | ... | Larvae move very actively and constantly |

* 100 larvae in each experiment.

† Heading "Length of Exposure" applies to experiments *D*, *E*, *E*1, *F*, *F*1, *G*, *G*1.

‡ "Moist" indicates that water was applied as required to maintain a fairly constant amount of moisture inside petri dish.

‖ "Dry" indicates that no water was applied to filter paper inside petri dishes after experiment was begun.

¶ Dead at end of exposure.

and 12 per cent dead pupae. Thus these conditions failed to stimulate pupation, for under field conditions when larvae were forced to pupate on the soil surface they did so in from 24 to 70 hours.

At temperatures of 90° to 100° F the mortality was very high and in proportion to the temperature and length of exposure. A relatively small percentage of larvae pupated at 90° F in moist tests while none pupated in the dry tests, nor in either the moist or dry tests at 100° F. Temperatures of 115° and 125° F for ½ hour were fatal to a high percentage of the larvae, while at a ¾-hour exposure, all larvae were killed. All temperatures between 90° and 125° F to which larvae were exposed greatly stimulated activity, and in these tests the larvae were in motion constantly.

## PUPA

*Time Required for Various Stages.*—The following experiment was conducted in an effort to obtain desired data on pupation. At the peak of larval emergence from the nuts, 12 battery jars were filled to a depth of 3 inches with sifted top soil from an orchard. The soil was packed in a standard manner and the battery jars were then buried to a depth of 3 inches in the soil surrounding a walnut tree. This arrangement very closely approached orchard conditions. One hundred mature larvae were taken directly from host tissue and were dropped onto the soil in each container, which was then covered with cheesecloth. In order to determine the necessary time for the puparia to be formed, single jars were removed at 6-hour intervals and the soil sifted. If puparia had not formed, the material was discarded after noting the degree of development. This was necessary in order to eliminate possible error by disturbing the larvae before they became completely inactive. Within 12 hours a few larvae were beginning to contract in length, assume a straw color, and otherwise show indications of the formation of the puparium. Within 18 hours, approximately 75 per cent of the larvae were in this condition; and in 24 hours the puparium was completely formed in practically all instances.

In entering the pupal stage the plastic larval skin hardens. This phenomenon takes place progressively from the posterior end forward. A decided straw color becomes evident with the hardening. The segments of the larvae are contracted and the anterior three segments are telescoped into the fourth. This leaves the anterior spiracles projecting slightly forward.

After the time required for formation of the puparium was determined, the subsequent morphologic changes were studied. The time required under field conditions for the consummation of these various

changes was also determined. Batches of 50 puparia of known age were carefully dissected at 6-hour intervals and the stage of development recorded. These records were tabulated and the development described, employing the terminology used by Snodgrass[36] in his study of the anatomy and metamorphosis of *Rhagoletis pomonella.* Within 36 hours after the formation of the puparium, there is an additional larval molt. The larva is enclosed within a hard shell and consequently remains stationary. This condition is commonly referred to as the prepupal stage. Histologic and morphologic changes continue, and within 90 to 100 hours after the puparium is formed the insect is in the early phase of the cryptocephalic stage. At this time small rudiments of the developing appendages are evident, though the future head is not visible. Within 10 to 20 hours more the final phase of the cryptocephalic stage has been reached. The rudiments of the appendages are larger than in the early phase, the abdomen still retains its prepupal form, no head is visible, and the prepupal skin has been shed over the entire body.

After 10 to 20 hours more have elapsed, the early phase of the phanerocephalic stage is reached. The head is everted though relatively small in size, the appendages are of increased size, and the abdomen still retains its prepupal form. The second phase of the phanerocephalic stage, or the final pupal stage, is attained within 10 to 12 hours more. The general form of the adult body is recognizable. The head is very large and the appendages are about full length, extending nearly to the end of the abdomen. Thus under the prevailing field conditions, which were fairly representative, the true pupal stage (fig. 11, p. 374) was reached within 145 to 175 hours after the larva entered the soil.

*Population of Pupae in Soil.*—In 1929, in this same grove just prior to the beginning of emergence, representative areas of soil were sifted under two trees in order to estimate the total number of pupae present. There was an average of 26 pupae per square foot of soil under tree No. 1, and the total area directly beneath the tree was 1,134 square feet. Thus the calculated total number of pupae was 29,484. Under tree No. 2 there was an average of 31 pupae per square foot and the total area was 1,385 square feet. Thus the calculated number of pupae present was 42,935.

*Soil Temperature as a Mortality Factor.*—In adult emergence studies of 1930, an attempt was made to determine the effect on time and rate of emergence when pupae were under natural conditions in soil in cages exposed to total sunlight in contrast to partial shade. Accordingly, in the fall of 1929, areas for emergence cages were seeded after the method described previously. Two cages were in total sunlight in the grove, while the other two were in the usual locations under trees in partial

TABLE 21

MORTALITY OF PUPAE AT VARYING DEPTHS IN SOIL UNDER TOTAL SUNLIGHT AND PARTIALLY SHADED CONDITIONS

| Cage No. | Location | State of pupae | Per cent of total pupae at various depths in soil | | | | | | Total | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | 0–1 inch | 1–2 inches | 2–3 inches | 3–4 inches | 4–5 inches | 5–6 inches | Number | Per cent |
| 8 | Total sunlight | Alive | 0.0 | 1.0 | 1.0 | 0.2 | 0.4 | 0.0 | 13 | 3 |
| | | Dead | 61.0 | 22.0 | 10.0 | 3.0 | 2.0 | 0.2 | 478 | 97 |
| 8a | Total sunlight | Alive | 0.0 | 0.0 | 8.0 | 3.0 | 0.0 | 0.0 | 15 | 11 |
| | | Dead | 24.0 | 34.0 | 27.0 | 4.0 | 0.7 | 0.0 | 127 | 89 |
| 9 | Total shade | Alive | 0.4 | 12.0 | 43.0 | 11.0 | 3.0 | 1.0 | 197 | 69 |
| | | Dead | 0.4 | 10.0 | 17.0 | 4.0 | 0.7 | 0.0 | 88 | 31 |
| 9a | Partial shade | Alive | 0.0 | 16.0 | 34.0 | 2.0 | 9.0 | 2.0 | 63 | 65 |
| | | Dead | 9.0 | 14.0 | 7.0 | 2.0 | 1.0 | 1.0 | 34 | 35 |

shade. When seasonal adult emergence began in 1930, the cages in total sunlight yielded no flies. Several weeks after emergence had begun, a few of the pupae in the soil of these two cages were examined and found to be dead. It was then desirable to obtain more detailed information. A total of 4 square feet of soil in different locations in each cage was carefully excavated in layers of 1 inch in thickness, and sifted. The pupae obtained were determined to be alive or dead by crushing, which is a reliable test. Data resulting from these excavations are presented in

TABLE 22

DISTRIBUTION OF PUPAE WITH RESPECT TO DEPTH IN ORCHARD SOIL DUE TO CULTIVATION PRACTICES

| Lot No. | Number of pupae per sq. ft. at various depths in soil | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Top | 0–1 in. | 1–2 in. | 2–3 in. | 3–4 in. | 4–5 in. | 5–6 in. | 6–7 in. | 7–8 in. | 8–9 in. | 9–10 in. | 10–11 in. | 11–12 in. | 12–14 in. |
| 1 | 0 | 3 | 5 | 1 | 3 | 2 | 2 | 1 | 0 | 1 | 0 | 0 | 0 | 0 |
| 2 | 0 | 4 | 2 | 2 | 4 | 7 | 6 | 3 | 5 | 3 | 2 | 4 | 0 | 0 |
| 3 | 1 | 0 | 2 | 1 | 0 | 0 | 4 | 3 | 0 | 0 | 0 | 0 | 0 | 0 |
| 4 | 0 | 6 | 2 | 1 | 3 | 10 | 2 | 10 | 5 | 2 | 0 | 2 | 0 | 0 |
| 5 | 0 | 1 | 3 | 7 | 3 | 11 | 7 | 0 | 5 | 1 | 1 | 1 | 3 | 0 |
| 6 | 0 | 0 | 0 | 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| 7 | 0 | 0 | 0 | 3 | 1 | 3 | 0 | 2 | 1 | 1 | 0 | 0 | 0 | 0 |
| 8 | 1 | 1 | 3 | 2 | 5 | 1 | 1 | 6 | 6 | 3 | 7 | 2 | 1 | 0 |
| 9 | 0 | 4 | 0 | 7 | 2 | 3 | 1 | 5 | 2 | 2 | 0 | 1 | 3 | 0 |
| 10 | 0 | 2 | 1 | 4 | 7 | 0 | 1 | 2 | 3 | 2 | 0 | 0 | 3 | 0 |
| Total | 2 | 21 | 18 | 30 | 28 | 37 | 24 | 32 | 27 | 15 | 10 | 10 | 10 | 264 |
| *Per cent* | *1* | *8* | *7* | *11* | *11* | *14* | *9* | *12* | *10* | *6* | *4* | *4* | *4* | *100* |

table 21. The data indicate that apparently a high mortality results from high soil temperatures when the pupae remain undisturbed in the soil in total sunlight.

Normal orchard-cultivation practices alter the situation somewhat. In late June, 1928, after the last cultivation of the soil, before emergence began and before soil temperatures became high, the relative distribution of pupae with respect to depth was studied. The grove in which the studies were made is the same one referred to as grove A in table 18 and also the one from which the data presented in table 22 were obtained. One square foot of soil was sifted from each of ten locations within the grove. Strata 1 inch in thickness were sifted individually. The results are presented in table 22.

As a result of cultivation more than half of the pupae were fairly evenly distributed at depths of from 3 to 8 inches, while an appreciable number were found at a depth of 12 inches. Thus cultivation of the soil serves to decrease environmental resistance considerably.

*Moisture and Dryness as Mortality Factors.*—The only information available regarding the effect of moisture and dryness on pupal mortality is forthcoming from general observations over a period of years. Occasionally an "indicator" adult emergence cage was located on soil that was very wet for periods of weeks at a time when the pupae were in the soil. At the other extreme, cages were sometimes inadvertently placed on very dry soil. Although detailed records of emergence from these indicator cages were not kept, many flies emerged, which demonstrates that neither the condition of excess moisture nor that of dryness was fatal to all of the pupae. In many instances adults have emerged from pupae kept indoors in glass vials without soil for a year. In one instance, under these conditions, adults emerged two years after the larvae pupated.

*Summary of Pupal Mortality.*—The extent of pupal mortality was fairly well determined over a period of three years for pupae of varying age. Normal pupae of known age were used in these studies. In all instances the pupae of different ages were kept segregated. The data obtained from these studies are presented in table 23. It is evident that the percentage of mortality varies greatly among pupae of different ages and in different seasons.

TABLE 23

MORTALITY OF PUPAE UNDER NATURAL CONDITIONS*

| Year | Age of pupae | Total number pupae | Mortality |
|---|---|---|---|
| | *years* | | *per cent* |
| 1929 | 1 | 2,000 | 39 |
| 1930 | 1 | 2,000 | 51 |
| | 2 | 1,400 | 33 |
| | 3 | 35 | 17 |
| 1931 | 1 | 2,000 | 23 |
| | 2 | 657 | 56 |
| | 3 | 54 | 85 |
| | 4 | 5 | 60 |

*Pupae buried in appropriate containers at depth of 5 inches under walnut trees.

*Dormancy.*—The nature of dormancy, or the diapause phenomenon, in *Rhagoletis completa* is very interesting though apparently complicated. A brief treatment of this phenomenon in insects was published in 1931,[6] in which reference to *R. completa* was made and certain limited data presented. However, since that time more information has accumulated and the total data have been reworked. The data presented

previously in the treatment of various factors affecting adult emergence (pp. 404–425) indicate that under natural conditions temperature is a very important physical factor relating to the matter.

To summarize briefly, the calculated seasonal peak of emergence was reached approximately 27 days earlier, and the percentage of annual-generation flies was increased approximately 26 per cent after the mild winters over that after cold winters. Also the time of emergence was earlier in those cases where the soil received the greatest heat from sunlight throughout the dormant period of annual-generation individuals. However, the exact relation of temperature to dormancy is not known beyond circumstantial evidence, which indicates that a certain minimum amount of heat units supplied by a range of fluctuating temperatures seem to be required for the termination of dormancy. Under fairly constant temperature conditions in the laboratory, dormancy was not broken to any appreciable extent when the equivalent number of total day-degrees temperature apparently required under field conditions had elapsed. It seems probable that in some manner temperature, together with unknown genetical factors, regulates the length of the dormant period in multi-annual-generation individuals. A summary of certain features pertaining to dormancy has already been presented in table 2 (p. 420).

Limited studies were conducted to determine the effect upon dormancy of varying lengths of exposure to temperatures of 20° and 30° F. One hundred pupae were used in each experiment. All experiments were begun on the same day. Pupae 20 days old were taken directly from soil and placed in moist sand in glass containers in which they were subjected to the several temperature conditions. The control lot was kept in an indoor insectary, and likewise all other lots at the termination of the respective periods of exposure to low temperatures. The data obtained are presented in table 24.

In only a few instances was there any indication that dormancy was materially altered by the treatments. In tests 3, 4, 5, 6, and 8 there were material increases in percentage of total emergence at the end of 7 months, when compared with the control. In these same tests, with the exception of test 6, increases in percentage of total emergence were yet evident at the end of 12 months, in comparison with the control. In general, the mortality at the end of 12 months was slightly less in the treated series than in the control. Repeated tests are necessary before important conclusions are warranted.

Preliminary experiments were conducted in an effort to determine the effect of certain chemicals in terminating the diapause. Only a few of the chemicals that have given positive results in breaking the dor-

mancy of plants were tested. In each test 200 pupae of the same age were used. Random concentrations of the materials were used, with several variations. Treatment consisted of soaking the pupae for varying lengths of time in the liquid, or subjecting them to an atmosphere of gas in certain instances. After treatment each lot was segregated in moist sand in containers that were kept indoors under heated insectary conditions. Adequate controls were maintained. Emergence of flies was recorded at weekly intervals. The materials and concentrations used and length of exposure, together with results obtained, are summarized in table 25.

TABLE 24

EFFECT OF TEMPERATURE UPON DORMANCY OF PUPAE*

| Experiment No. | Temperature | Exposure | Emergence 7 mos. | Emergence 12 mos. | Mortality 12 mos. | Pupae alive 12 mos. |
|---|---|---|---|---|---|---|
| | | | Per cent of total pupae | | | |
| | *degrees F* | *hours* | | | | |
| 1 | 20 | 33 | 0 | 22 | 50 | 28 |
| 2 | | 47 | 5 | 19 | 57 | 24 |
| 3 | | 154 | 10 | 48 | 38 | 14 |
| 4 | | 240 | 14 | 47 | 26 | 27 |
| 5 | | 336 | 11 | 46 | 43 | 11 |
| 6 | | 792 | 24 | 31 | 63 | 6 |
| 7 | 30 | 312 | 4 | 42 | 19 | 39 |
| 8 | | 528 | 40 | 50 | 39 | 11 |
| 9 | | 1,084 | 8 | 28 | 54 | 18 |
| 10 | 73 | Continuous control | 4 | 36 | 51 | 13 |

* 100 pupae were used in each experiment.

Potassium thiocyanate apparently produced a slight effect upon dormancy, since an appreciable percentage emerged within 138 days after treatment. The mortality, however, was materially higher than in comparable controls after 335 days. Thiourea appeared to stimulate emergence somewhat after 130 days without any effect upon the mortality. Ethylene chlorohydrin as gas, ethylene dichloride, carbon bisulfide, hydrocyanic acid gas, and xylene were fatal to the pupae under the conditions tested. The results obtained in these studies, particularly with potassium thiocyanate and thiourea, though inadequate to base conclusions upon, suggest that these and other chemicals might profitably be employed in further studies of the nature of dormancy in insects.

TABLE 25

EFFECT OF CERTAIN CHEMICALS UPON DORMANCY OF PUPAE*

| Material | Experiment No. | Concentration: Liquid, per cent | Concentration: Gas, cc vaporized per 10,000 cc space | Exposure, hours | 15 days | 60 days | 130 days | 166 days | 190 days | 335 days | 335 days: Per cent mortality | 335 days: Per cent alive |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | Accumulated emergence in per cent of total pupae | | | | | | | |
| Ethylene chlorohydrin | 1 | ........ | 2.5 | 24 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 |
| | 1a | ........ | 5.0 | 24 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 |
| | 1b | 5.0 | ........ | 1 | 0 | 0 | 5 | 10 | 10 | 15 | 61 | 24 |
| | 1c† | 5.0 | ........ | 1 | 3 | 4 | 7 | 20 | 21 | 29 | 59 | 12 |
| | 1d | 10.0 | ........ | 1 | 1 | 1 | 2 | 11 | 11 | 13 | 66 | 21 |
| | 1e | 20.0 | ........ | 1 | 0 | 1 | 3 | 22 | 25 | 35 | 44 | 21 |
| Ethylene dichloride | 2 | ........ | 2.0 | 24 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 |
| Potassium thiocyanate | 3 | 5.0 | ........ | 1 | 0 | 0 | 2 | 24 | 30 | 37 | 49 | 14 |
| | 3a | 10.0 | ........ | 1 | 1 | 2 | 4 | 39 | 56 | 62 | 35 | 3 |
| Ammonium thiocyanate | 4 | 5.0 | ........ | 1 | 0 | 0 | 2 | 18 | 23 | 37 | 45 | 18 |
| | 4a | 10.0 | ........ | 1 | 0 | 0 | 2 | 9 | 10 | 17 | 71 | 12 |
| Thiourea | 5 | 5.0 | ........ | 1 | 1 | 2 | 6 | 25 | 35 | 53 | 20 | 27 |
| | 5a | 10.0 | ........ | 1 | 2 | 4 | 4 | 31 | 43 | 56 | 26 | 18 |
| Carbon bisulfide | 6 | ........ | 1.0 | 24 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 |
| | 6a | ........ | 2.0 | 24 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 |
| Carbon tetrachloride | 7 | ........ | 2.5 | 24 | 0 | 0 | 1 | 1 | 1 | 2 | 98 | 0 |
| Hydrocyanic acid | 8 | ........ | 2.5 | 24 | 0 | 0 | 0 | 0 | 0 | 0 | 100 | 0 |
| Xylene | 9 | ........ | 2.5 | 24 | 0 | 0 | 0 | 0 | 1 | 2 | 98 | 0 |
| Control | 10 | Distilled water | ........ | 1 | 0 | 1 | 3 | 21 | 25 | 38 | 19 | 43 |
| | 11† | Distilled water | ........ | 1 | 4 | 4 | 5 | 17 | 18 | 23 | 64 | 13 |
| | 12 | No treatment | ........ | .... | 1 | 1 | 5 | 17 | 19 | 30 | 28 | 42 |

* Two hundred pupae were used in each test. All pupae were 45 days old when treated, unless otherwise indicated.

† Pupae one year old.

## SEASONAL HISTORY

Host resistance and accumulated soil-temperature conditions during dormancy apparently exert a profound effect upon seasonal activity of *Rhagoletis completa.* Pertinent facts regarding seasonal history for the five-year period of this study are summarized in figure 57.

*Adult Emergence.*—Official weather records within the infested area indicate that winter temperatures preceding the 1928 season and temperatures during that season were slightly warmer than normal for this period; however they more nearly approached normality than in any following year during this five-year study. The calculated median of adult emergence (time when 50 per cent of total emergence had occurred) was August 17. The number of days elapsed from the time 50 per cent of the larvae pupated was 321. Accumulated temperature totaled 19,232 day-degrees, and the monthly departure from normal averaged +1.7 degrees. Approximately 70 per cent of the pupae that were formed in 1927 emerged in 1928 and are therefore classed as annual generation. The emergence curve, if smoothed, may be considered a normal frequency polygon, indicating normality of emergence.

In 1929 the median of adult emergence occurred on August 24, and the total period that pupae remained in the soil was 328 days, or 7 days longer than in 1928. The total day-degrees of temperature was 19,286, only 55 degrees more than in 1928, and the monthly departure from normal was –13.8 degrees. Approximately 45 per cent of the 1928 pupae constituted the annual generation. The emergence curve was of the same general shape as that of 1928. In comparing the emergence in 1928 with that of 1929, the data indicate that in 1929 the median was delayed as a result of temperature conditions during dormancy of the pupae.

In 1930 the median of adult emergence occurred on July 29, and the total period that pupae remained in the soil was 306 days, which was 22 day's shorter than in 1929, and 15 days shorter than in 1928. The total day-degrees of temperature was 18,313, and the monthly departure from normal was +3.6 degrees. Approximately 90 per cent of the 1929 pupae constituted the annual generation. The emergence curve in 1930 is altered in shape materially from that of previous seasons. The multimodal effect was probably the result of temperature conditions during dormancy. If time of emergence is assumed to be regulated solely by accumulated soil temperature, other factors being equal, then these data would indicate that a slight increment of temperature greatly accelerates emergence.

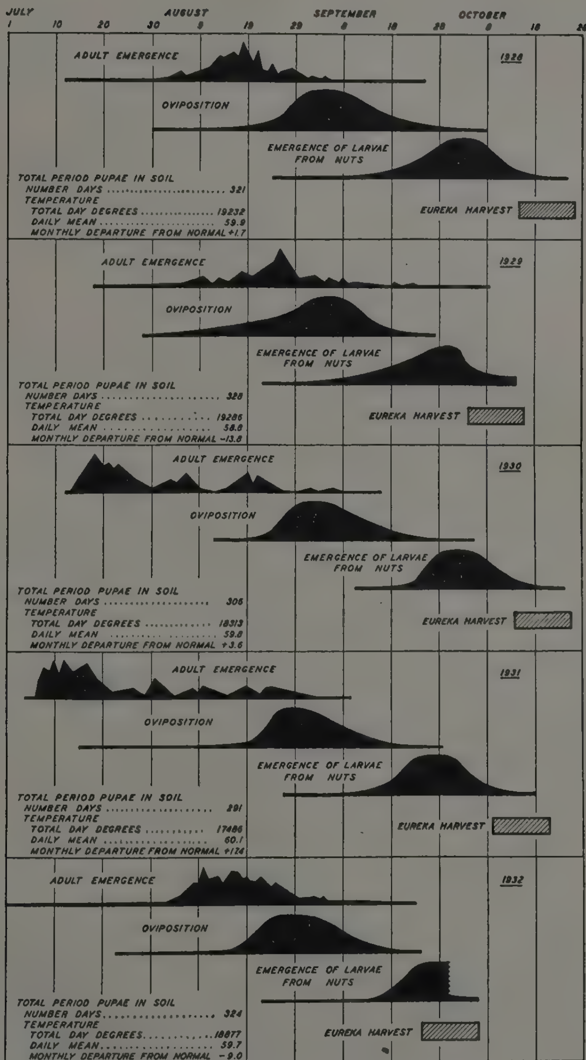



Fig. 57. Seasonal history of *Rhagoletis completa* in the Chino-Pomona area (1928–1932).

In 1931 the median of adult emergence was reached on July 18, and the total period that pupae remained in the soil was 291 days. Thus emergence was 11 days in advance of the previous earliest season, and 25 days in advance of the latest season. The total day-degrees of temperature was 17,486; and the monthly departure from normal was +17.4 degrees. Approximately 83 per cent of the 1930 pupae constituted the annual generation. The shape of the emergence curve probably indicates the operation of abnormal conditions.

At the end of the 1931 season, after four years of study, it became a matter of conjecture whether the early emergence in 1930 and 1931 was wholly due to abnormally high seasonal temperatures or whether the insect was becoming synchronized with the milder climate of this state. The climate of the region in which it is indigenous, particularly the winter season, is rigorous, and adults emerge considerably later in the year than in the latest season here. The matter was economically important here; for, should the flies emerge during the middle of June, it is probable that all varieties of walnuts would be susceptible to infestation.

In 1932 the median of adult emergence occurred on August 16, and the total period that pupae remained in the soil was 324 days. Thus emergence was 29 days later than in 1931, and 1 day earlier than in 1928, which was more nearly normal than any other season during these studies. The accumulated day-degrees of temperature was 18,877, or 355 degrees less than the elapsed day-degrees in 1928, and the monthly departure from normal was −9.0 degrees. Approximately 67 per cent of the 1931 pupae constituted the annual generation. The shape of the 1932 emergence curve approached normality, again resembling that of the 1928 and 1929 seasons.

In consideration of the available information, it appears that an unknown combination of temperatures during dormancy is the most important physical factor governing the time of emergence and also the percentage of individuals that constitute the annual generation. The 1928 and 1929 data indicate that under the existing fluctuating temperature conditions prevailing during the period of dormancy, when a certain amount of day-degrees of temperature was accumulated, transformation to adulthood was initiated. This suggests that the transformation to adulthood represents the completion of physiological processes that for the most part are dependent upon a combination of temperature conditions peculiar to insects with this type of diapause. Continuing this line of reasoning further, the 1930 and 1931 data possibly indicate that these physiological processes are consummated in a shorter period of time and with less actual heat units when the temperatures are higher

during the warm phase of the fluctuating temperature cycle. On this hypothesis the 1932 data indicate that the insect actually is becoming acclimatized, for with subnormally low temperatures during dormancy, transformation to adulthood occurred in practically the same number of days as in 1928, and with fewer day-degrees temperature. Should these generalizations be founded on facts, it follows that in normal seasons the median of adult emergence would probably be reached early in August instead of during the middle of August, which was the case during the earlier history of the insect in its new habitat.

*Oviposition Period.*—Extensive data regarding hardness of the green walnut husk indicate that this physical factor is the most important of all relating to varietal susceptibility as well as time of oviposition in a susceptible variety. It is interesting to note (fig. 57) that the peak of oviposition each year for the five-year period was reached between August 29 and September 5, despite the wide variation in time of emergence. In each season except 1929 the ascending slope of the oviposition curve was relatively steep, indicating, in 1930 and 1931 particularly, that an inhibitory factor lost its effect rather suddenly. To visualize the situation, females were vainly attempting to oviposit and were not successful until the husk softened sufficiently to permit insertion of the ovipositor. The green husks increase in hardness from the time they are formed until the latter part of June, when the peak of hardness is reached. They gradually soften with approaching maturity. The husk apparently reaches a susceptible condition for oviposition on nearly the same calendar date each year. Should this feature of host susceptibility become an established fact, after a few years of further observation, it would greatly reduce the expense incident to control measures, since one properly timed application would probably be satisfactory. Furthermore the time for treatment would probably become a calendar date, thereby ignoring the emergence of flies for practical purposes of treatment.

*Emergence of Larvae from Nuts, and Time of Harvest.*—It is evident from figure 57 that during seasons when harvest did not begin before October 10, the greater percentage of larvae had emerged prior to harvest. In 1929 the walnuts ripened early with the result that many contained immature larvae when harvested. The 1932 harvest was the earliest encountered in these studies. Less than half of the larvae had emerged from the nuts and many failed to mature. However, in most instances the larvae had developed sufficiently to cause the shell of the nut to become stained; therefore the extent of injury and resultant loss was not materially mitigated.

## NATURAL ENEMIES

*Rhagoletis completa* is remarkably free from important natural enemies. The total effect of the activities of those species that do attack this insect apparently does not increase environmental resistance sufficiently to constitute an important factor in the economy of the species.

*Fungi.*—Several species of pathogenic fungi that bring about a mortality among adults under laboratory conditions have been recorded. These species are: *Metarrhizium anisopliae, Botrytis bassiana, Entomophthora* sp., and *Cladosporium* sp. (determined by Charles). These fungi have been found only on flies confined in the battery-jar cages. Occasionally all larvae inhabiting a walnut are found dead in the husk tissue, apparently as a result of a fungal or bacterial pathogen. *Fusarium* sp. has been cultured from these larvae in several instances.

*Arachnoid Species.*—The mite *Pediculoides ventricosus* New. (determined by Ewing) has been occasionally observed feeding upon the eggs of the walnut husk fly, inside the egg cavity. In all instances a single mite was present and had consumed more than half of the eggs. The mite had entered the cavity through the small puncture made by the female in ovipositing but was so engorged when observed that it occupied nearly one-half of the cavity. Apparently they do not become abundant enough to reduce the numbers of their host materially.

The following spiders, determined by Banks, have been observed preying upon either larvae or adults of the walnut husk fly: *Agelena pacifica* Bks., *Epeira prompta* Htz., *Epeira* sp., *Aysha decepta* Bks., *Zelotes* sp., *Icius vitis* Ckll., *Tetragnatha laboriosa* Htz., *Psilochorus apicalis* Bks., *Pardosa sternalis* Thor., *Ebo* sp., and another *Ebo* sp. which is probably undescribed. While immature and adult spiders utilize appreciable numbers of larvae and adults for food, they are of minor importance in the control of the fly.

*Hexapod Species.*—Nymphs and adults of the anthocorid, *Triphleps insidiosus* (Say) (determined by Van Duzee), have been commonly observed feeding upon the eggs of the fly. They insert their beaks into the egg cavity through the hole in the husk made for oviposition. In the instances observed, dissection of the cavity immediately after their feeding showed that less than one-half of the eggs present had collapsed. Detailed studies previously reported indicated that approximately 12 per cent of egg mortality at that time could be attributed to the work of natural enemies, probably by this bug and the *Pediculoides* mite. The reduviid, *Zelus renardi* Koln., has commonly been noted with an adult fly impaled on its beak.

The chrysopid, *Chrysopa californica* Coq., and particularly the larvae as they approach maturity, prey upon adults of the walnut husk fly whenever they succeed in capturing them.

Three species of ants (determined by Smith) have been observed preying upon either larvae or adults of the fly. A large grayish species, *Formica cinerea* subsp. *pilicornis* Emery, captures adults as they emerge from the soil and also on the foliage, particularly at night when the flies do not move about to any appreciable extent. This ant also has been observed while transporting larvae to its nests. The tiny thief ant, *Solenopsis molesta* var. *validiuscula* Emery frequently raided the cages in the field laboratory until isolation in water was practiced. They destroyed adults, larvae, and pupae. The tiny black ant, *Monomorium minimum* Buck., destroyed appreciable numbers of larvae in the husks of black walnuts. These nuts usually drop to the ground early in the season before the larvae reach maturity; consequently larval development continues within the walnut lying on the soil.

Throughout the course of these investigations, which involved thousands of larvae and pupae, special efforts have been made to determine the existence of parasites. None has been observed. H. S. Smith conducted a special search for parasites of the larvae in Kansas, the region in which the species is undoubtedly indigenous. He found none; apparently not even the dipterous larval parasites of general feeding habits attack this species. However, two general-feeding parasites of dipterous pupae, the chalcid *Spalangia rugosicollis* Ash. and the proctotrupid, *Galesus* sp., very near *atricornis* Ash. (determined by Gahan), were reared from *Rhagoletis completa* pupae at Manhattan, Kansas, in 1931. They appear to be of negligible importance.

Through the coöperation of the United States Bureau of Entomology, Smith introduced into California from Hawaii the opiine larval parasites *Opius humilis* Silv. and *Diachasma tryoni* Cam. Both species were reared under laboratory conditions from *Rhagoletis completa*. Field colonizations were made in 1931 and 1932 and *O. humilis* was recovered in 1932; however to date *D. tryoni* has not been taken in the field.

*Other Animals.*—Fowls in general, particularly chickens, and birds eat the larvae and pupae; but their activities are limited and are unimportant.

## SCAVENGER SPECIES INHABITING DECAYING WALNUT HUSKS

More than 30 species of insects with scavenger habits have been found associated with or immediately following *Rhagoletis completa* larvae in the decaying walnut husk. Many of those recorded are dipterous larvae from which adults have been reared. Only a few of the more common ones have been determined; they are: *Euxesta putricola* Cole (determined by Cole), *Lonchaea occidentalis* Mall., *Muscina assimilis* Fall, *Fannia canicularis* (Linn.) (all determined by Aldrich), and *Drosophila* spp. The first two species are usually present in all decaying walnut husks, though in certain seasons one species may become more abundant than the other. There is a continuous breeding of these scavenger flies. Late in the season, after walnut husk fly larvae have emerged, larvae of various sizes and comprising several species, are usually abundant.

Several species of beetles have been recorded, though *Carpophilus hemipterus* (Linn.) is most common. A staphylinid species, predacious upon the dipterous scavenger larvae present, usually became abundant late in the season.

The scavenger species increase the husk injury caused by the walnut husk fly; however, their presence seldom appreciably increases the economic loss to the producer.

## CONTROL STUDIES

Preliminary life-history data at the beginning of these studies clearly indicated that the adult stage is the most vulnerable to mechanical control practices. The major portion of this phase of the study has therefore been devoted to determining the relative merits of the more promising available toxic materials for destroying the adult and also determining their effect on the walnut tree.

Acid lead arsenate has been used since 1912 in controlling the apple maggot, *Rhagoletis pomonella* (Walsh) and the cherry fruit flies, *R. fausta* (O.S.) and *R. cingulata* (Loew). It was also used against the Mediterranean fruit fly, *Ceratitis capitata* Wied. in the eradication campaign in Florida. But since walnut foliage is very susceptible to injury from acid lead arsenate, this material was eliminated from consideration for field trials. The basic type of lead arsenate does not cause injury and is used extensively on walnuts for the control of the codling moth, *Carpocapsa pomonella*. At the beginning of this study, basic lead arsenate was the only known promising insecticide used as a stomach

poison that could be applied to walnut foliage with safety. However, there was uncertainty as to the expected efficacy of it in comparison with acid lead arsenate since it contains approximately 30 per cent less arsenic oxide ($AS_2O_5$) with generally a reduction of the amount that is water-soluble.

The control studies are treated under two categories, namely, laboratory investigations and field investigations. The laboratory work extended from 1929 to 1932, while the field work began in 1928 and extended through 1932. Since these investigations were conducted concurrently with the life history and other biological studies, the combined data of any one year somewhat influenced the procedure of the following year.

## LABORATORY TOXICOLOGICAL INVESTIGATIONS

Reliable information concerning the toxic effect of basic lead arsenate and other materials on the walnut husk fly was necessary in order to guide future field trials. Therefore experiments were undertaken to obtain this information. They consisted in testing materials under comparable conditions to determine the relative effectiveness of each as measured by the speed of fatality. Some difficulty was experienced in working out a satisfactory technique, particularly with reference to such factors as suitable food and cages for favorable conditions for the flies, rapid handling of the flies in fairly large numbers without inflicting injury, application of standard amounts of materials to be tested, and maintenance of host material bearing toxic elements in a satisfactory condition to insure significant results. The methods devised and used for conducting these studies served the purpose fairly satisfactorily, though they were not refined to a degree suitable for highly accurate toxicological studies such as the determination of mean lethal concentrations. All tests were conducted in a shaded, screened laboratory located in a walnut grove.

*Procedure.*—Walnut twigs of the Eureka variety, bearing two nuts each, were used as the unit of each test. An effort was made to secure nuts of uniform size for each series. They were taken from unsprayed trees. The twig was cut at a point 8 or 10 inches back from the nuts and immediately placed in water. Each nut was washed in several changes of tap water, as were the bottles into which they were subsequently placed. With bottles partially filled with water, several inches of the stem were cut off and the remaining portion bearing the nuts was placed in the bottle and allowed to dry in the sun. Then absorbent cotton was tightly packed around the stem to support it in an upright position. The cotton also served to prevent the flies from becoming trapped in the water.

It was necessary to provide the flies with some form of food, since repeated tests earlier had shown that they die very rapidly when confined on walnuts under cage conditions. Therefore a stock solution containing 10 grams of granulated cane sugar to 100 cc of tap water was used as the liquid portion of the mixture to be atomized onto the nuts. In the 1931 tests the amount of sugar used was increased to 20 grams. Most of the inorganic stomach poisons were tested at a concentration of 1:100. (Concentrations of each material tested are indicated in the charts summarizing results.) Approximately 5 cc of the mixture was atomized onto each unit of two nuts with a De Vilbiss atomizer from a standard distance, the atomizer being shaken continuously during the application. Controls received stock sugar solution only, and in amounts equal to those received by other units of the series.

Where the materials were applied as a dust, various concentrations were used ranging from 20 to 90 per cent, by weight, in the regular tests, to 100 per cent in certain special tests. The remaining portion of the dust mixture consisted of from 10 to 50 per cent powdered cane sugar plus the necessary percentage of diluent to bring the mixture up to 100 per cent. Hydrated lime was used as a diluent for the arsenical and copper compounds; while talc, diatomaceous earth, and bentonite were used for the fluorines and other materials. Application was made with a De Vilbiss dust applicator. Approximately 1 gram of the dust mixture was applied to the unit of two nuts. Some controls in the dusted series received an application of powdered cane sugar, while those used to determine whether or not the diluent material effected mortality were treated with a mixture of the particular material and 10 or 20 per cent powdered cane sugar.

The walnuts were placed in the inverted battery-jar cage, one unit to each cage (fig. 58) immediately after treatment in the dust tests, and as soon as thoroughly dry in the liquid tests. However, in certain tests involving the employment of nicotine or other volatile materials, 16-mesh screen cages were used to obviate a fumigating effect. These cages were similar in size and shape to the battery-jar cages and were handled in a like manner. Flies were confined in the cages to feed upon the treated walnuts. Controls were maintained for each test variation.

In 1931 tests were conducted to determine the effect of certain materials applied as dusts directly onto the bodies of the flies. No dilution of materials was made in these instances. The flies used in each test were placed in a large test tube, 2 inches in diameter. The nozzle of the De Vilbiss dust applicator was inserted through the cotton plug stoppering the tube and a uniform amount of material was blown into the tube. The amount was sufficient to fill the tube with a cloud of dust particles. In

this manner flies occupying the tube were contacted by the dust particles which adhered to their bodies in a fairly uniform manner. As soon as the dust cloud settled, the flies were removed and placed in the inverted battery-jar cage containing a cluster of two nuts bearing a coating of stock sugar solution. Two sets of controls were included in each series of these tests: in one powdered cane sugar was dusted onto the insects and in the other the regular operation was simulated except that

Fig. 58. Inverted battery-jar cage and typical set-up used in toxicity studies.

air void of dust materials was blown into the tube. These tests will be referred to in future discussion as "contact" series.

The flies were handled in the manner previously described. In tests within a series they were usually the same age, though in some cases there was a difference of 1 day. In no instance were the flies over 4 days old when the experiment was set up. Approximately equal numbers of each sex were used. Generally from 20 to 50 flies were used in each test, according to the number available at the time the series was started. However, the number was practically constant within each unit of a series. Flies used were kept in stock cages for 1 or 2 days after they had emerged from the soil, and those injured during collection from the emergence cages were not included in the toxicity series. Since all tests within a series were set up on the same day, and under comparable conditions, the daily mortality rate was regarded as an index of the relative merits of each material tested. The number of dead flies in each test within the regular series was recorded three times daily, while those in the contact series were recorded at 2-hour intervals.

*Interpretation of Data.*—From a study of literature pertaining to insect toxicology it is apparent that a satisfactory comparison of the toxicity of insecticides at given concentrations can be made when the average time required for 50 per cent mortality is used as a standard. Relative to this matter, Fisher, according to Tattersfield and Morris,[37] states:

> The relation between concentration and probability of death could theoretically be determined by experiment by exposing a large number of insects to the action of the insecticide at each concentration. However, the number of insects required increases enormously if we wish to explore in this manner the region in which the probability of death is high. If as many as 99 per cent of the insects were killed, the accuracy of the comparison between any two insecticides would depend upon the comparatively few insects which survived; and to compare them with any accuracy many thousands of insects would have to be used. The same difficulty arises in the comparatively unimportant case when the deaths are few. For a given number of insects the most accurate comparison can be made when the concentrations are such that about 50 per cent perish. The region between 25 per cent and 75 per cent can be fairly easily explored. It is for this reason that the preliminary examination of chemical substances should be made by comparison of the concentrations required to give a mortality of 50 per cent. When the equivalent at this point is established it would be most valuable to ascertain if the same relative concentrations are equivalent over the range 25–75 per cent. Only in this way does it seem possible to infer a general equivalence of insecticidal properties. The direct comparison of mortality when the probability of survival is very small would seem to be beyond the scope of accurate laboratory investigation.

Bar diagrams are employed in figures 59, 62, 65, and 69 to present the data obtained from these studies, using the average time required for 50 per cent mortality as the standard of comparison.

Figures 60, 61, 63, 64, 66, 67, and 68 show in more detail the relative effectiveness of the materials tested, the standard of comparison being the average time required for mortality of from 25 to 75 per cent. Plotted on semilog paper, these points fall fairly well along a straight line. Throughout this range the data indicate the regular sigmoid toxicity curve relation.[38] The curves have not been mathematically fitted, since the conclusions to be drawn from the data do not appear to warrant such detailed treatment.

*Experiments in 1929.*—Data obtained from laboratory studies of the effectiveness of the various materials tested in 1929 are presented in figures 59, 60, and 61.

The arsenicals tested were effective in causing mortality of the flies. These tests show that the speed of toxic action bears a direct relation to the arsenic content and degree of solubility in water. Basic lead arsenate applied as dust was more rapid in its action than when applied as spray

| 1929 MATERIAL AND CONCENTRATION | | HOW APPLIED | NUMBER TESTS | TOTAL NUMBER FLIES |
|---|---|---|---|---|
| CONTROL | 10% SUCROSE | SPRAY | 4 | 114 |
| | POWD. SUCROSE | DUST | 4 | 111 |
| CALCIUM FLUORIDE | 1:100 | SPRAY | 3 | 70 |
| CA. FLUOSILICATE COMPOUND | 1:100 | SPRAY | 3 | 59 |
| BASIC LEAD ARSENATE | 1:100 | SPRAY | 3 | 73 |
| (LIME AS DILUENT) | 20% | DUST | 3 | 74 |
| ACID LEAD ARSENATE | 1:100 | SPRAY | 3 | 51 |
| COPPER SULFATE | 1:100 | SPRAY | 3 | 132 |
| BARIUM FLUORIDE | 20% | DUST | 3 | 58 |
| BARIUM FLUOSILICATE | 1:100 | SPRAY | 2 | 53 |
| (TALC AS DILUENT) | 20% | DUST | 2 | 59 |
| SODIUM FLUORIDE | 1:100 | SPRAY | 1 | 20 |
| (TALC AS DILUENT) | 20% | DUST | 2 | 39 |
| MG. FLUOSILICATE | 1:100 | SPRAY | 2 | 37 |
| SODIUM FLUOSILICATE | 1:100 | SPRAY | 3 | 72 |
| (TALC AS DILUENT) | 20% | DUST | 4 | 96 |
| CALCIUM ARSENATE | 1:100 | SPRAY | 2 | 45 |
| SODIUM ARSENITE | 1:100 | SPRAY | 2 | 40 |
| NICOTINE SULFATE | 5:100 | SPRAY | 2 | 71 |
| | | TOTALS | 51 | 1274 |

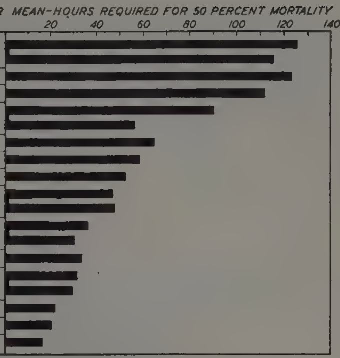



Fig. 59. Results of toxicity studies in 1929, showing number of hours required to bring about 50 per cent mortality of *Rhagoletis completa.*

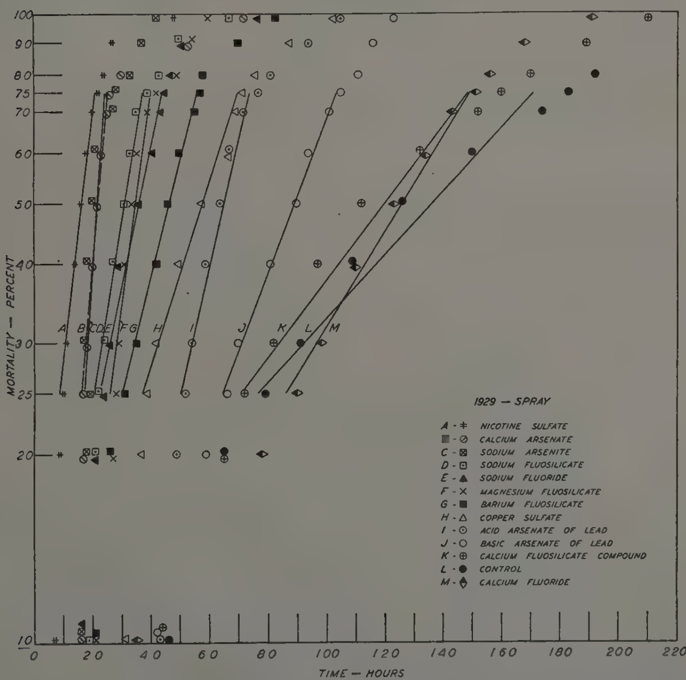



Fig. 60. Relative effectiveness of various materials applied as spray in 1929. Standard of comparison is the mean mortality period from 25 to 75 per cent.

and even more rapid than the acid lead arsenate applied as spray. Since comparable amounts of these materials, by weight, were applied, perhaps the dust particles are more readily ingested by the flies. Furthermore previous work had established the fact that more material was retained by the surface of the walnut in dust tests than in spray tests, and

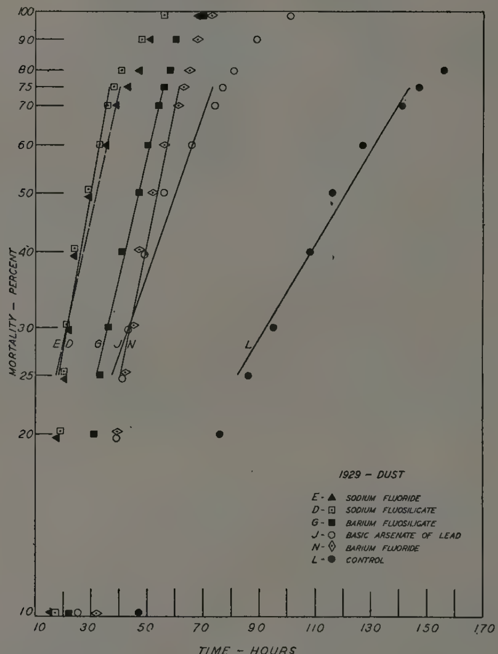



Fig. 61. Relative effectiveness of various materials applied as dust in 1929. Standard of comparison is the mean mortality period from 25 to 75 per cent.

the amount retained, both of basic and acid lead arsenates, adheres to plant surfaces more firmly when applied as spray than when dusted.

All fluorines tested, except those combined with calcium, killed the flies fairly rapidly. The speed of toxic action is correlated with the solubility of the material in water, the more soluble compounds being more rapid in their lethal action. Calcium fluoride and calcium fluosilicate compound were not effective, which fact may possibly be corroboratory evidence regarding the theory of action of the fluorines in bringing

about insect mortality:[26] that they kill by virtue of the affinity of fluorine for calcium, the fluorine precipitating calcium in the body tissues, thereby interfering with permeability. On this basis a possible conclusion is that the affinity of the fluorine in the calcium combinations tested is already satisfied, partially or wholly, thereby rendering them ineffective. However, the solubility of the calcium combinations tested is very low, which fact alone may account for the low insecticidal efficiency.

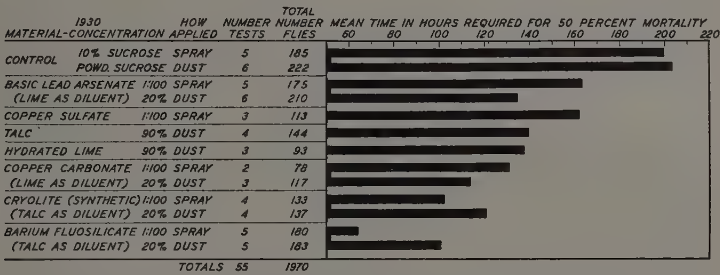



Fig. 62. Results of toxicity studies in 1930, showing number of hours required to bring about 50 per cent mortality of *Rhagoletis completa*.

Nicotine sulfate was employed as a stomach poison. The sugar of the stock food solution apparently reduced the volatilization of the nicotine. The action of nicotine in these tests was the most rapid of the materials tested.

Copper sulfate was more rapid in its action than acid lead arsenate. The nature of the lethal action of copper is not understood; it may kill directly by poisoning, or indirectly by destroying the intestinal microorganisms.

Tree-tolerance tests in 1929 eliminated acid lead arsenate, calcium arsenate, sodium arsenite, sodium fluoride, sodium fluosilicate, and magnesium fluosilicate from further trials except for comparative toxicity purposes, since important injury resulted from their use. Calcium fluoride and calcium fluosilicate compound were eliminated because of low insecticidal efficiency. Barium fluoride was not continued since it was less practicable and no more efficacious than barium fluosilicate.

*Experiments in 1930.*—Data obtained from laboratory studies of the effectiveness of various materials tested in 1930 are presented in figures 62, 63, and 64. The speed of toxic action of certain materials was considerably slower in 1930 than in 1929. This variable longevity factor is likewise evident when a comparison is made of the length of life in the

controls for both seasons. The explanation of these differences is not known. Materials and technique were identical in both seasons. However, the performance of materials used both seasons and the controls show a fairly close ratio of variation, i.e., basic lead arsenate required twice as long to bring about 50 per cent mortality in 1930 as in 1929, and

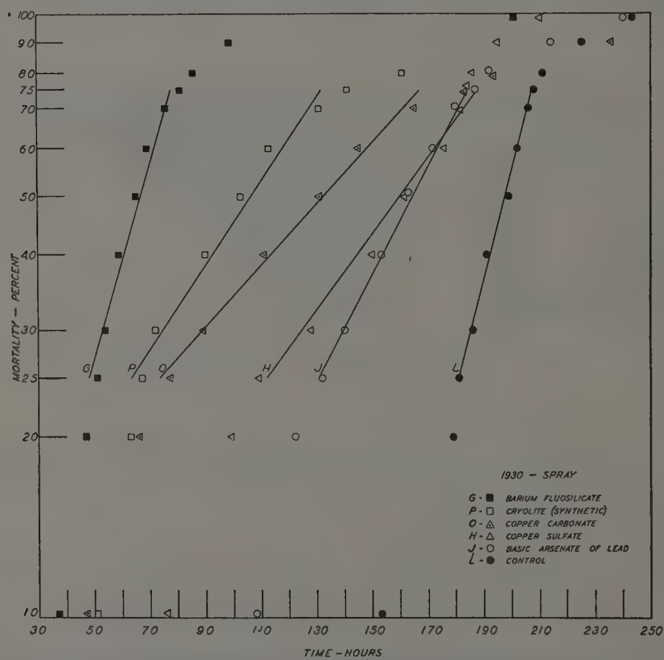



Fig. 63. Relative effectiveness of various materials applied as spray in 1930. Standard of comparison is mean mortality period from 25 to 75 per cent.

controls lived about 1.6 times as long in 1930 as in 1929. Climatic conditions may have been the influencing factor; temperature and humidity control was not undertaken in any of these studies.

Basic lead arsenate, applied as dust, exhibited more rapid action than the spray, as in 1929.

Copper sulfate was not as effective as in 1929, and copper carbonate surpassed the sulfate form in speed of toxic action. Here again the dust was more rapid than the spray in killing the flies. Both materials were eliminated from further trials because of injury to walnut foliage in field trials.

Of the fluorines tested, barium fluosilicate was superior to synthetic cryolite (sodium fluoaluminate). With both materials the spray tests showed a more rapid killing effect than dust tests.

Talc and hydrated lime both exhibited insecticidal action. The mortality rates were closely approximate to each other and also to that of

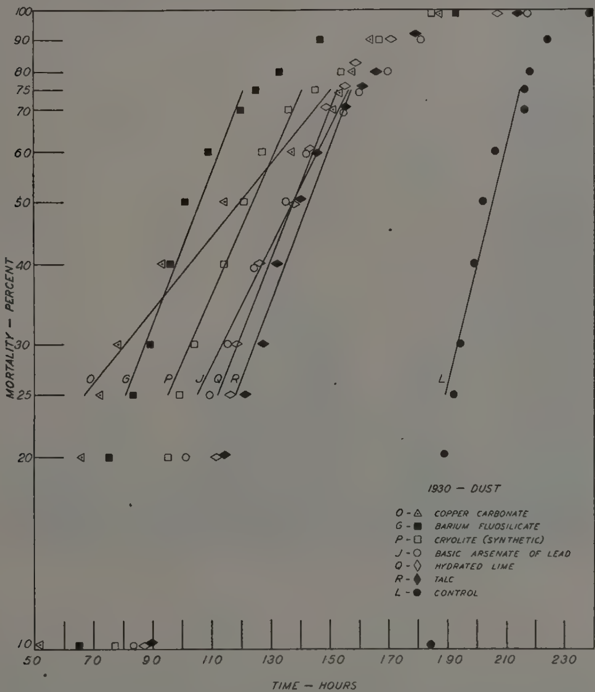


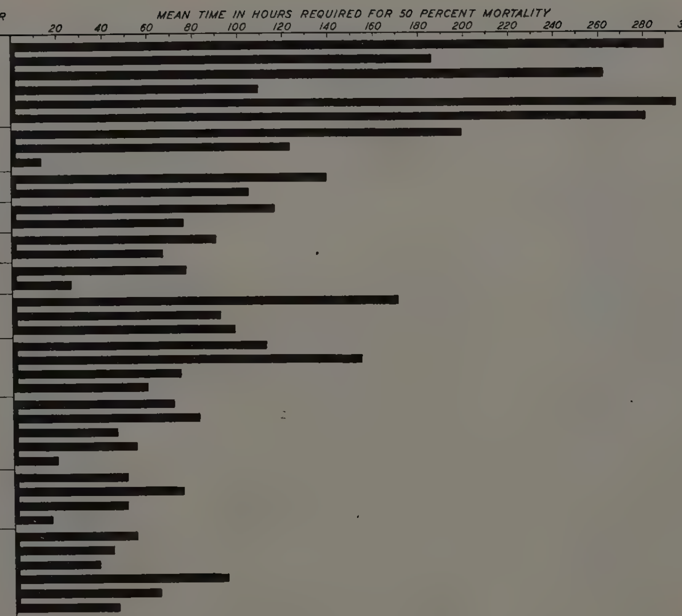

Fig. 64. Relative effectiveness of various materials applied as dust in 1930. Standard of comparison is mean mortality period from 25 to 75 per cent.

basic lead arsenate applied as a dust. The nature of action of talc and hydrated lime is not understood.

*Experiments in 1931.*—Data obtained from laboratory studies of the effectiveness of various materials tested in 1931 are presented in figures 65, 66, 67, and 68.

The controls in 1931 show that the inverted battery-jar cages afford more favorable conditions for longevity than do the screen cages. Furthermore when the food (sucrose) is supplied as a spray the flies live

| 1931 MATERIAL AND CONCENTRATION | | HOW APPLIED | NUMBER TESTS | TOTAL NUMBER FLIES |
|---|---|---|---|---|
| CONTROLS | GLASS CAGE-20% SUCROSE | SPRAY | 3 | 102 |
| | SCREEN CAGE-20% SUCROSE | SPRAY | 3 | 107 |
| | GLASS CAGE-POWD. SUCROSE | DUST | 4 | 138 |
| | SCREEN CAGE-POWD. SUCROSE | DUST | 2 | 88 |
| (CONTACT)[1] | GLASS CAGE-POWD. SUCROSE | DUST | 4 | 96 |
| (CONTACT) | GLASS CAGE-POWD. SUCROSE | DUST | 6 | 160 |
| DIATOMACEOUS EARTH | 80% +12% WHITE OIL | DUST | 2 | 58 |
| | 80% | DUST | 3 | 97 |
| | (CONTACT)[1] 100% | DUST | 5 | 128 |
| TALC | 80% | DUST | 2 | 61 |
| | (CONTACT)[1] 100% | DUST | 4 | 102 |
| SULFUR | 80% | DUST | 2 | 59 |
| | (CONTACT)[1,2] 100% | DUST | 4 | 100 |
| BENTONITE | (CONTACT)[1] 100% | DUST | 4 | 102 |
| | 80% | DUST | 2 | 56 |
| HYDRATED LIME | 80% | DUST | 2 | 57 |
| | (CONTACT)[1] 100% | DUST | 4 | 96 |
| BASIC LEAD ARSENATE (LIME AS DILUENT) | 1:100 | SPRAY | 2 | 47 |
| | 20% | DUST | 2 | 47 |
| | 40% | DUST | 2 | 60 |
| CRYOLITE (NATURAL) (TALC AS DILUENT) | 1:100 | SPRAY | 2 | 49 |
| | 20% | DUST | 2 | 42 |
| | 40% | DUST | 2 | 59 |
| | (CONTACT)[1] 100% | DUST | 4 | 100 |
| BARIUM FLUOSILICATE (TALC AS DILUENT) | 1:100 | SPRAY | 2 | 62 |
| | 20%+12% FISH OIL | DUST | 2 | 56 |
| | 20% | DUST | 2 | 55 |
| | 40% | DUST | 2 | 63 |
| | (CONTACT)[1] 100% | DUST | 4 | 103 |
| CRYOLITE (SYNTHETIC) (TALC AS DILUENT) | 1:100 | SPRAY | 2 | 59 |
| | 20% | DUST | 2 | 67 |
| | 40% | DUST | 2 | 60 |
| | (CONTACT)[1] 100% | DUST | 4 | 100 |
| NICOTINE SULPHATE-TANNIC ACID | 1-3:500[2] | SPRAY | 2 | 67 |
| NICOTINE SULPHATE-BUTANOL-WHITE OIL | 1-1-15:500[2] | SPRAY | 2 | 71 |
| NICOTINE SULPHATE-SUCROSE | 1-50:500[2] | SPRAY | 2 | 64 |
| NICOTINE 2% IN BENTONITE[2] | 80% | DUST | 2 | 54 |
| NICOTINE 2% IN DIATOM. EARTH[2] | 80% | DUST | 2 | 66 |
| NICOTINE 2% (GROUND TOBACCO)[2] | 80% | DUST | 2 | 70 |
| | | TOTALS | 106 | 3028 |




NOTE
[1] MATERIAL APPLIED DIRECTLY ON BODIES OF INSECTS
[2] SCREEN CAGES USED

Fig. 65. Results of toxicity studies in 1931, showing number of hours required to bring about 50 per cent mortality of *Rhagoletis completa*.

longer in screen cages than when powdered sugar is dusted onto the nuts. Powdered sucrose applied directly to the bodies of the flies had no deleterious effect upon them.

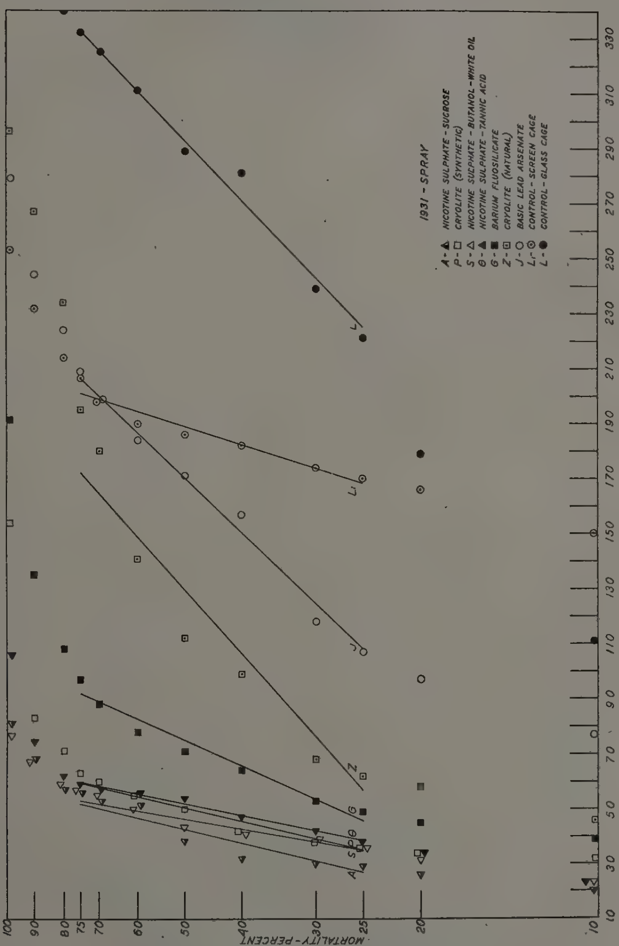



Fig. 66. Relative effectiveness of various materials applied as spray in 1931. Standard of comparison is mean mortality period from 25 to 75 per cent.

In the contact series the mortality caused by the contact of the materials with the bodies was very probably an indirect effect, since they undoubtedly ingested relatively large amounts of the material as a result of their "cleaning-up" habits. For several hours after treatment

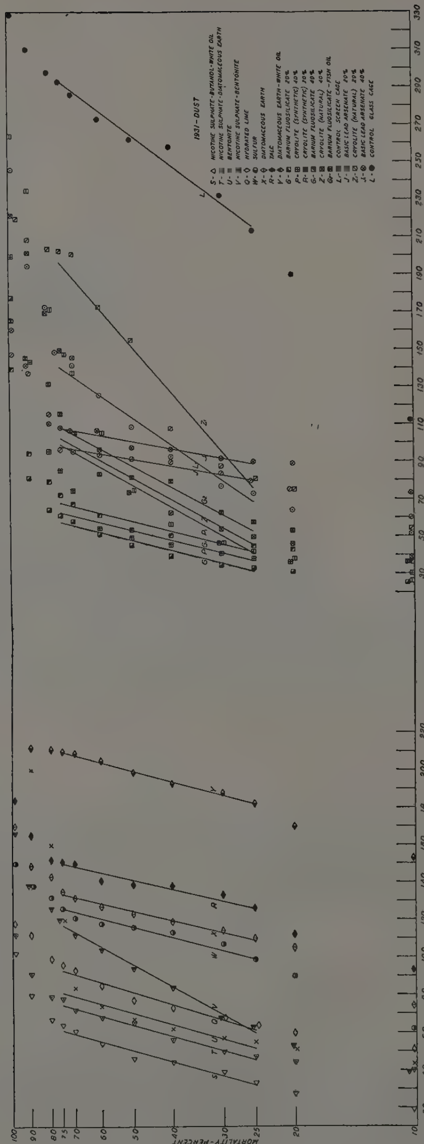



Fig. 67. Relative effectiveness of various materials applied as dust in 1931. Standard of comparison is mean mortality period from 25 to 75 per cent.

they were observed to be constantly removing the dust from their bodies with their legs, the fore pair usually being cleaned with the mouth parts.

Of all materials used, diatomaceous earth was the most rapid in its lethal action when applied directly onto the bodies of the flies. It pro-

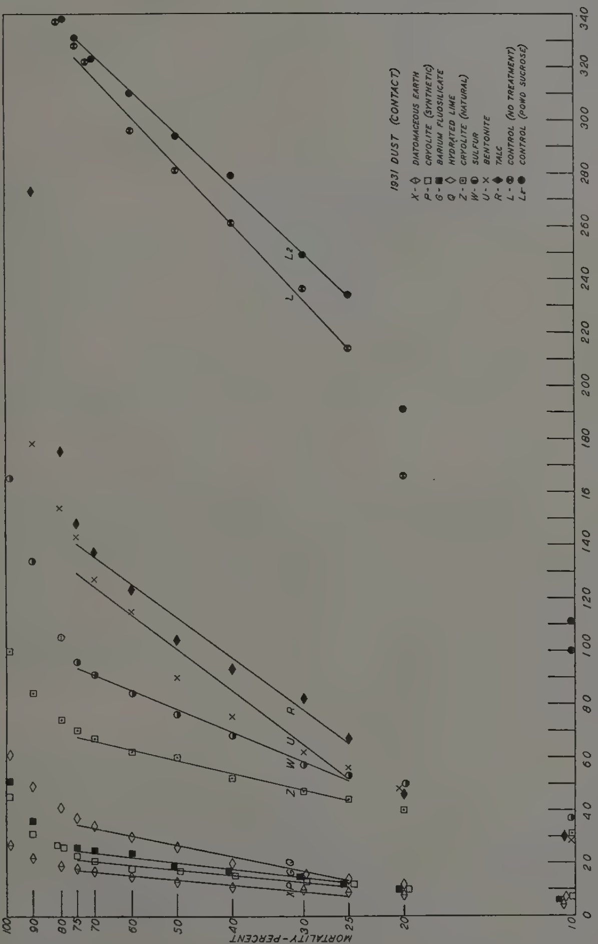



Fig. 68. Relative effectiveness of various materials applied as dust directly onto the bodies of the insects in 1931. Standard of comparison is mean mortality period from 25 to 75 per cent.

duced mortality much more slowly when flies fed upon it normally, and still more slowly when mineral oil was incorporated therein. Oil causes the earth particles to adhere more firmly to the surface of the nut,

thereby apparently rendering it more difficult for the flies to pick up in feeding. Talc, sulfur, bentonite, and hydrated lime were also more rapid in their action when applied directly onto the bodies of the flies than when flies fed normally upon them. Apparently larger amounts of material are ingested during the removal of the material from their bodies than through normal feeding.

All diluent materials used, in dusts applied to the nuts, that is, diatomaceous earth, talc, sulfur, bentonite, and hydrated lime, demonstrated lethal action upon the flies, the latter being generally more rapid than the others. The nature of this action is unknown; however, the following theories are suggested, although they are highly problematical. Diatomaceous earth and talc may seriously abrade the intima and epithelium, and possibly other tissues of the alimentary canal. Furthermore, they may produce a diarrheal condition as a result of the cathartic action of magnesium, the content of which is relatively high. Sulfur may produce a highly acid condition in the digestive system, thereby causing deleterious metabolic disorders. Bentonite is colloidal in nature and apparently withdraws moisture from the intestinal walls; however, instead of the usual flushing action, there is a clogging of the rectum as a result of increased volume due to water adsorption. Therefore, death may ensue as a result of failure to eliminate excrement. Hydrated lime may produce a somewhat hardened layer on the inner lining of the intestine, resulting in a failure to digest and assimilate food, thereby causing mortality through starvation, which may likewise be an important factor in mortality produced by diatomaceous earth, talc, and sulfur.

Considering the known toxicity of nicotine, the nicotine compound and mixtures tested were relatively slow in their action as stomach poisons. The differences evidenced in speed of action of the various mixtures possibly indicate the degree to which the nicotine is bound within or by the respective compounds and mixtures.

Basic lead arsenate exhibited relatively slow action; and spray tests were slower in producing mortality than were dust tests. The performance of this material in 1931 was in accord with the 1929 and 1930 tests. The 40 per cent dust mixture was somewhat slower in action than the 20 per cent mixture. Considering the speed of action of hydrated lime, it is possible that the increased amount of this diluent material in the 20 per cent lead arsenate mixture was responsible for the observed difference in speed of action over the 40 per cent lead arsenate mixture.

Barium fluosilicate and synthetic cryolite (sodium fluoaluminate) on the whole exhibited similar speeds of toxic action. Fish oil incorporated in the barium fluosilicate mixture appreciably retarded the effect of the latter. The explanation of this fact is presumably that sug-

gested previously for the diatomaceous earth and mineral oil mixture. Observation showed that the fish oil was not repellent to the flies. In the barium fluosilicate dust tests, the 40 per cent mixture was slower in its action than the 20 per cent mixture, which fact is not readily explained. In speed of action, natural cryolite was considerably inferior to the synthetic material in these tests. Similar results were obtained by Ripley and Hepburn[34] in toxicity studies dealing with the Natal fruit fly, *Pterandus rosae* (Ksh.). The two materials are identical in composition and the ones used in these tests were similar in physical properties. Pos-

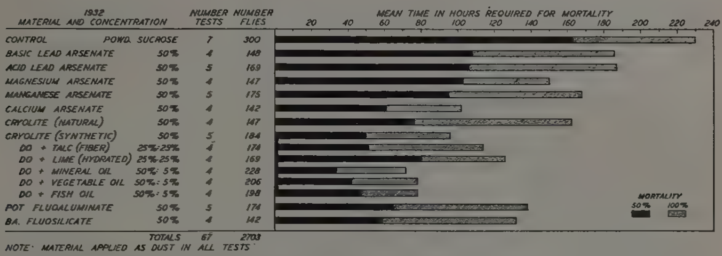



Fig. 69. Results of toxicity studies in 1932, showing number of hours required to bring about 50 per cent and 100 per cent mortality of *Rhagoletis completa*.

sibly the synthetic material is more effective because the bond between the elements of the molecule is not as great as in the natural product, thereby liberating the fluorine more readily.

*Experiments in 1932.*—Data obtained from laboratory studies of the effectiveness of various materials tested in 1932 are presented in figure 69.

The longevity of flies in the controls was sufficient to attribute significance to the indicated data from tests involving the use of toxic material.

Magnesium arsenate was employed in a field control plot in 1932, while manganese arsenate received consideration in tree-tolerance tests. It was therefore important to determine the relative effectiveness of these two materials in comparison with other arsenicals and fluorine compounds. The data indicate insignificant differences in the speed of action of basic lead arsenate and acid lead arsenate, with slightly increased efficacy for manganese and magnesium arsenate, and greatest rapidity of action for calcium arsenate.

Natural cryolite was more effective than any of the arsenicals tested with the exception of calcium arsenate. However, it was slower in action than any of the other fluorine compounds tested.

Synthetic cryolite exhibited greatest speed of toxic action of all materials tested. Regarding the effect on speed of action when a calcium diluent is used, the data indicate appreciable inhibition. An inert diluent such as talc apparently exerts no effect upon toxic action. The 1931 studies showed that the incorporation of 12 per cent mineral oil or fish oil in dust mixtures for adhesive purposes materially reduced the rate of mortality. Further information regarding this matter was desirable, therefore tests were conducted involving the use of three types of oil. Preliminary studies of mineral oil, vegetable oil, and fish oil as adhesives had shown that satisfactory results were obtained when a 5 per cent concentration of any one was employed. These toxicity studies indicate increased speed of action in the tests where mineral oil and vegetable oil were employed. The differences shown appear to be significant, particularly in the mineral-oil test; however, for the purpose of this investigation the most important consideration is that none of the oils used inhibited toxic action.

Barium fluosilicate and potassium fluoaluminate are highly efficacious materials. The physical properties, particularly the bulkiness of the latter, are superior to other fluorine compounds tested.

*Ingestion Studies.*—In control studies dealing with insects possessing mouth parts of the sponging and sucking type, the nature of lethal action of certain solid materials with low solubility in water is not definitely understood. Researchers on the *Rhagoletis pomonella* problem have opined that lead arsenate is not ingested by the flies, since the particles are "strained out" of imbibed liquid by the pseudotracheae or other structures on the labella. Thus, in order for the material to cause death, it would be necessary for the flies to ingest appreciable quantities of arsenic in solution. Considering the water-solubility of commercial lead arsenate, it seems improbable that lethal amounts could be obtained in this manner. However, quantities of the material may be readily dissolved in the saliva which the flies emit in normal feeding and which is subsequently imbibed.

In feeding studies on nonbiting flies, Graham-Smith[18] found that the blow fly (*Calliphora erythrocephala* Mg.) readily ingests all particles measuring up to 0.006 mm in diameter, and that particles measuring up to 0.02 mm in diameter are drawn into the pseudotrachea when two opposite interbifid grooves are made to communicate with each other. Furthermore, relatively large objects apparently pass directly into the mouth when the prestomal cavity is open during prolonged sucking efforts made by the flies. Thus these studies show that the structure of this type of mouth part permits ingestion of solid matter. The mouth parts of the blow fly are somewhat similar in structure and size

to those of the walnut husk fly. For enlightenment on this matter, an effort was made to determine whether or not flies actually ingested the solid particles of lead arsenate. In preliminary tests lead arsenate was colored with aniline blue, methyl green, orange *G*, and säurefuchsin, all of which are water-soluble stains. Individual tests were conducted in which the dyed material was dusted onto walnuts and flies caged on them. After several days' confinement under such conditions, dissection showed that the gut of most of the flies contained material similar in color to that with which the lead arsenate was originally dyed. Furthermore, excrement of the color used was present on the sides of the battery-jar cage. This indicated that the flies had ingested the stained particulate matter; however, definite evidence was lacking since only the water-soluble stain itself may have been taken in. Therefore similar tests were conducted in 1929 and 1930, in which non-water-soluble stains were used. Of the many tested, Sudan *III*, which is soluble in alcohol, served the purpose most satisfactorily. A saturated solution of the stain was made and thoroughly mixed with a small amount of the lead arsenate (approximately 20 grams), thus making a paste. This was allowed to dry at laboratory temperature. When completely dry, the material was pulverized and the procedure repeated. After three or four successive mixings with the dissolved stain, the lead arsenate was the color of the stain, scarlet red.

Lead arsenate, barium fluosilicate, cryolite, copper carbonate, hydrated lime, talc, diatomaceous earth, bentonite, sulfur, and ground tobacco leaves were stained in the manner just described. Individual cage tests of each material applied on walnuts as spray and dust were conducted. In these tests the materials were used undiluted, except for the 20 per cent sucrose that was included for food. Controls were maintained in which the walnuts were treated with sucrose only. Fifty flies were used in each test. Approximately 48 hours after the flies were confined on the treated walnuts, lots consisting of 25 flies from each test were removed, stupefied with ether, and dissected. All dissection was performed with the aid of a binocular microscope at a magnification of $\times 50$. In all tests over 98 per cent of the flies dissected had large quantities of stained material present in various portions of the alimentary canal; however, it was not abundant in the hind intestine. Varying amounts were also present on the labella of the proboscis. In the controls, the alimentary canal of all flies dissected was creamy white, except in certain instances where the rectum presented a slightly brownish coloration.

Just prior to stupefaction for dissection, when the vial containing the flies was held so as to permit light to pass through the abdomen, the in-

testine containing stained material was plainly visible in all instances. This was not observed in any of the flies from control cages. In all tests, except where stained bentonite was employed, characteristic reddish excrement was present on the sides of the battery-jar cage. The recta of most of the flies consuming bentonite were greatly distended and upon further dissection proved to be effectively plugged with a somewhat gum-like material. Apparently this colloidal substance had absorbed relatively large amounts of moisture, and greatly increased volume had resulted.

In the diatomaceous-earth tests, microscopic examination of stained contents from various portions of the alimentary canal revealed the presence of diatome shells. Furthermore these minute shells were found in the stained excrement taken from the battery-jar cage of this particular test. These observations demonstrate conclusively that particulate matter is ingested by the flies, while the evidence from the tests involving other materials is considered to be strongly indicative of this fact. Undoubtedly the flies imbibe these undissolved particles in suspension in droplets of dew, or in their own saliva, which is emitted in the process of feeding.

*Comments on Toxicological Investigations.*—Walnut husk flies are favorable subjects for toxicological studies of this nature, since the death point is readily determined. Furthermore the type of cage used permitted accurate observations on the rate of mortality.

Cage tests of this nature are not actual comparisons of the toxicity of the various materials, as has been pointed out by Campbell.[10] The insects feed freely and consume unknown quantities of the material. However, such tests are deemed satisfactory for comparing the relative effectiveness of materials when such factors as those of biological and environmental nature are comparable. It is recognized that these studies have not been extensive enough or sufficiently well controlled to warrant important toxicological conclusions. A possible partial explanation of erratic results obtained in some instances may be attributed to the time of beginning a series of experiments in relation to the time and the amount of feeding of the flies just prior to being confined on the treated walnuts. Ideally, the flies should not have been permitted to feed at all until placed in the test cages. Thus they would all begin to consume the material at approximately the same time. However, this procedure was not feasible under existing conditions.

These studies have supplied valuable information for field plot trials. Basic lead arsenate has consistently demonstrated its relatively slow action in producing mortality, while barium fluosilicate and synthetic cryolite have been consistently efficacious in this respect.

## FIELD INVESTIGATIONS IN 1928

Two experiments were conducted in 1928, using basic lead arsenate. Because of the absence of summer rainfall, the loss of the arsenical from walnut foliage and fruit through weathering during the period of ac-

Fig. 70. Spraying equipment in operation in experimental control plots.

tivity of the fly is probably not very great. Therefore one coverage of this material, applied when the flies begin to emerge, should afford data regarding the efficacy of it in the control of the walnut husk fly.

*Plot Experiments I and II.*—A plot was sprayed (fig. 70) in each of two groves where the infestation was reported to have been appreciable

TABLE 26

CONTROL EXPERIMENTS IN 1928

| Experiment No. | Number of trees and variety | Material | Concentration | Date of application | Trees counted | Total nuts counted | Mean per cent infested† | Per cent control |
|---|---|---|---|---|---|---|---|---|
| I | (9½ acres) | | | | | | | |
| *A** | 43 Eureka | Basic lead arsenate | 4 lbs.-100 gals. | Sept. 1 | 4 | 831 | 28 | 71 |
| *B* | 119 Eureka | Check—no treatment | ........ | ........ | 4 | 762 | 96 | .... |
| II | (10 acres) | | | | | | | |
| *A** | 36 Eureka + 30 Neff | Basic lead arsenate | 4 lbs.-100 gals. | Sept. 2 | 6 | 1,334 | 25 | 65 |
| *B* | 84 Eureka + 70 Neff | Check—no treatment | ........ | ........ | 6 | 1,480 | 71 | .... |
| Totals | 4 plots, 382 walnut trees, 19½ | acres | ........ | ........ | 20 | 4,407 | .... | .... |

* One application of material as spray at 20 gallons per tree.

† Data obtained from count of nuts on lower portion of Eureka trees just before harvest (October 14).

in 1927. Each plot was approximately square in shape. The trees were not large and, therefore, 20 gallons of material per tree gave satisfactory coverage. Application was made on August 1 and 2. A few flies emerged in July; however, the daily number did not increase until early in August, reaching the seasonal peak on August 18 (fig. 36).

For evaluation of results counts were made of the percentage of infested nuts on representative trees in the centers of treated plots as well as in the adjoining check plots. The percentage of reduction in infestation or the percentage of control, was calculated after the method used by Porter.[31] $\overline{B}$ represents the mean percentage of infestation in the treated plot and $\overline{A}$ represents the mean percentage of infestation in the untreated plot. Then

$$100 - 100\frac{\overline{B}}{\overline{A}} = \text{per cent reduction in infestation or per cent control.}$$

A summary of the data obtained from these experiments is presented in table 26.

The data indicate that basic lead arsenate very materially reduced the degree of infestation. However, with 25 per cent infested nuts in treated plots the degree of control was not satisfactory. The season was apparently very favorable for the walnut husk fly; for heavy infestations developed in groves that were so lightly infested the preceding year as to be unnoticed.

## FIELD INVESTIGATIONS IN 1929

The performance of basic lead arsenate in 1928 was sufficiently encouraging to warrant further field trials; and since very little information was available concerning tree reaction from other materials, basic lead arsenate was the only toxic material tested in field control plots in 1929. The so-called "bait sprays" were given a considerable amount of attention. Plots were differentially treated with respect to concentration of materials, amount per tree, number of applications, and the timing of the applications.

It was regrettable that representative controls were not feasible during this season. Authorities were seriously considering an eradication campaign which prohibited the maintenance of any sizable untreated area. The "check" indicated in the several experiments was in all instances a border row contiguous to treated plots. Later studies have shown that such an arrangement does not yield truly significant results. Therefore, in attempting to evaluate the efficacy of the various treatments certain supplemental facts should be considered. The fly population within the experimental groves was sufficiently large to produce

serious damage in all instances had not lethal or other prohibitive factors been in operation. Reliable information regarding fly population was available from two sources. Emergence cages were in operation throughout the season within the respective groves. Furthermore sweetened material sprayed onto the tips of branches of representative trees served to congregate flies during their early-morning feeding activities. In this manner it was possible to note the relative abundance of flies within a grove.

Even though seasonal emergence records show that only approximately 45 per cent of the flies emerged, they were present in relatively large numbers. It is questionable whether or not the varieties infested in previous years were in a susceptible condition during the activities of the flies in 1929. It has been pointed out in the host studies that husk hardness appears to be the most important factor governing oviposition, and that this feature is variable with the same varieties in different groves. However, in several small untreated Eureka groves within the area, fairly heavy infestations developed. This information may or may not be significant in view of the lack of detailed data regarding such features as the ratio of nut population to female population and percentage of infestation.

In order to determine the degree of infestation in the various plots, fairly extensive counts of the nuts on the trees in the centers of the plots were made just prior to harvest. These counts included nuts distributed over all portions of the tree. Nuts were considered infested when eggs had been deposited in the husks.

Harvest was abnormally early. Therefore, an appreciable percentage of those nuts classified as "infested" in field counts failed to exhibit evidence of injury when harvested. Percentage of infestation does not necessarily signify percentage of injury; however, the former is the important consideration in evaluating the merits of the various materials tested.

Adult emergence was somewhat later than in 1928. A few flies emerged during the latter part of July and early August, with the seasonal peak being reached on August 26 (fig. 37, p. 409).

Experimental plots comprised approximately 70 acres and included practically all of the moderate to heavily infested properties in the area in 1928. A summary of these experiments is given in table 27.

*Plot Experiment I.*—Infested nuts were not found in any of the plots until the latter part of August. Flies were commonly observed in all parts of the grove throughout the entire season. The check trees were about one-half the size of the trees in the treated plots and were in an outer border row.

CONTROL EXPERIMENTS IN 1929

| Experiment | Number of trees and variety | Material | Concentration (per 100 gals. for sprays and per 100 lbs. for dusts) | Application: Method | Application: Am't per tree, gals. for sprays and lbs. for dusts | Application: Number | Application: Date | Foliage injury, "burn" | Estimated per cent infested 1928 | Results: Trees counted | Results: Total nuts counted | Results: Mean* per cent infested | Results: Per cent control* |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| I | (10 acres) | | | | | | | | | | | | |
| A | 77 Eureka | Basic lead arsenate | 4 lbs. | Spray | 20 | 3 | 7/20, 8/15, 9/4 | None | 70–90 | 6 | 2,262 | 0.4 | 94 |
| B | 77 Eureka | Basic lead arsenate | 25 lbs. | Dust | 1½ | 3 | 7/20, 8/15, 9/4 | None | 70–90 | 6 | 2,290 | 0.7 | 89 |
| | | Hydrated lime | 75 lbs. | | | | | | | | | | |
| C | 17 Eureka | Check, no treatment | ........ | ........ | .... | .... | ........ | ........ | 70–90 | 6 | 1,986 | 6.4 | .... |
| II | (5 acres) | | | | | | | | | | | | |
| A | 34 Eureka | Basic lead arsenate | 4 lbs. | Spray | 20 | 3 | 7/31, 8/16, 9/4 | None | 40–60 | 4 | 1,283 | 1.1 | 93 |
| B | 34 Eureka | Basic lead arsenate | 100 lbs. | Dust | 1 | 3 | 7/31, 8/16, 9/4 | None | 40–60 | 4 | 1,309 | 5.4 | 66 |
| C | 17 Eureka | Check, no treatment | ........ | ........ | .... | .... | ........ | ........ | 40–60 | 4 | 904 | 15.9 | .... |
| III | (10 acres) | | | | | | | | | | | | |
| A | 39 Eureka | Basic lead arsenate | 4 lbs. | Spray | 20 | 3 | 7/31, 8/15, 9/5 | None | 40–60 | 6 | 1,084 | 1.0 | 77 |
| B | 39 Eureka | Basic lead arsenate | 4 lbs. | Spray | 10 | 3 | 7/31, 8/15, 9/5 | None | 40–60 | 6 | 1,190 | 0.5 | 88 |
| C | 39 Eureka | Basic lead arsenate | 4 lbs. | Spray | 20 | 2 | 8/6, 8/25 | None | 40–60 | 6 | 1,071 | 0.1 | 98 |
| D | 39 Eureka | Basic lead arsenate | 4 lbs. | Spray | 10 | 2 | 8/6, 8/25 | None | 40–60 | 6 | 1,105 | 0.3 | 93 |
| E | 17 Eureka | Check, no treatment | ........ | ........ | .... | .... | ........ | ........ | 40–60 | 6 | 1,013 | 4.4 | .... |
| IV | (5 acres) | | | | | | | | | | | | |
| | 85 Eureka + 157 peach† | Basic lead arsenate | 4 lbs. | Spray | 15 | 3 | 8/2, 8/16, 9/4 | None | 20–40 | 6 | 1,694 | 30.1 | .... |
| V | (4 acres) | | | | | | | | | | | | |
| A | 34 Eureka | Basic lead arsenate | 4 lbs. | Spray | 20 | 3 | 8/1, 8/19, 9/7 | None | 10–20 | 5 | 1,062 | 2.4 | 46‡ |
| B | 34 Eureka | Basic lead arsenate | 4 lbs. | Spray | 20 | 3 | 8/1, 8/19, 9/7 | Slight | 10–20 | 5 | 1,051 | 4.4 | .... |
| | | Corn syrup "Karo" | 3 gals. | | | | | | | | | | |
| VI | (5½ acres) | | | | | | | | | | | | |
| A | 43 Eureka | Basic lead arsenate | 4 lbs. | Spray | 20 | 3 | 8/2, 8/19, 9/3 | None | 30–50 | 4 | 1,000 | 11.4 | 58 |
| B | 26 Eureka | Basic lead arsenate | 6 lbs. | Spray | 5 | 3 | 8/2, 8/19, 9/3 | Moderate | 30–50 | 4 | 1,139 | 13.9 | 49 |
| | | N. O. molasses | 5 gals. | | | | | | | | | | |
| | | Cane sugar | 25 lbs. | | | | | | | | | | |
| C | 26 Eureka | Check, no treatment | ........ | ........ | .... | .... | ........ | ........ | 30–50 | 4 | 934 | 27.0 | .... |

* Data obtained from count of nuts on Eureka trees (unless otherwise indicated) just prior to harvest (September 30).
† Peaches not treated. ‡ Indicates effectiveness of A over B.

TABLE 27—(*Concluded*)

| Experiment | Number of trees and variety | Material | Concentration (per 100 gals. for sprays and per 100 lbs. for dusts) | Application: Method | Application: Am't per tree, gals. for sprays and lbs. for dusts | Application: Number | Application: Date | Foliage injury, "burn" | Estimated per cent infested 1928 | Results: Trees counted | Results: Total nuts counted | Results: Mean* per cent infested | Results: Per cent control* |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| VII | (6 acres) | | | | | | | | | | | | |
| A | 85 Eureka | Basic lead arsenate<br>Table syrup§ | 6 lbs.<br>5 gals. | Spray | ½‖ | 3 | 7/31, 8/16, 9/4 | Moderate | 20–40 | 20 | 2,018 | 1.3 | 35 |
| B | 17 Eureka | Check, no treatment | ........ | ........ | .... | .... | ........ | ........ | 20–40 | 8 | 682 | 2.0 | .... |
| VIII | (1½ acres) | | | | | | | | | | | | |
| A | 13 Eureka | Basic lead arsenate | 8 lbs. | Spray | 15 | 2 | 8/4, 8/21 | None | 40–60 | 5 | 1,056 | 2.9 | 76‡ |
| B | 13 Eureka | Basic lead arsenate<br>Corn syrup "Karo" | 8 lbs.<br>5 gals. | Spray | 15 | 2 | 8/4, 8/21 | Moderate | 40–60 | 5 | 1,000 | 11.9 | .... |
| IX | (7½ acres) | | | | | | | | | | | | |
| A | 43 Eureka | Basic lead arsenate | 12 lbs. | Spray | 6 | 3 | 8/1, 8/18, 9/16 | None | 20–40 | 4 | 1,341 | 5.8 | .... |
| B | 85 Eureka | Basic lead arsenate<br>N. O. molasses<br>Beet sugar | 6 lbs.<br>5 gals.<br>25 lbs. | Spray | 3 | 3 | 8/1, 8/18, 9/16 | Moderate | 20–40 | 4 | 1,215 | 3.6 | 38** |
| X | (5 acres) | | | | | | | | | | | | |
| A | 42 Eureka<br>42 Franquette<br>1 Klondike | Basic lead arsenate<br>Corn sugar | 16 lbs.<br>25 lbs. | Spray | 3 | 2 | 8/5, 8/24 | Slight | 30–50<br>30–50<br>70–90 | 4<br>4<br>1 | 1,121<br>1,015<br>175 | 1.5<br>0.8<br>0.5 | ....<br>....<br>.... |
| XI | (10 acres) | | | | | | | | | | | | |
| A | 54 Eureka + 45 Neff | Basic lead arsenate<br>Corn sugar | 16 lbs.<br>25 lbs. | Spray | 3 | 3 | 8/3, 8/17, 9/4 | Slight | 40–60 | 6 | 2,307 | 6.8 | 75 |
| B | 54 Eureka + 45 Neff | Basic lead arsenate | 100 lbs. | Dust | 1¾ | 3 | 8/3, 8/17, 9/4 | None | 40–60 | 6 | 2,531 | 3.5 | 87 |
| C | 12 Eureka + 10 Neff | Check, no treatment | ........ | ........ | .... | .... | ........ | ........ | 40–60 | 6 | 2,114 | 27.0 | .... |
| | Totals—27 plots, 1,238 walnut trees, 157 peach, 70 acres | | | | | | | | | 161 | 38,952 | | |

* Data obtained from count of nuts on Eureka trees (unless otherwise indicated) just prior to harvest (September 30).

‡ Indicates effectiveness of A over B. § Probably made of unrefined sugar. ‖ Small trees. ** Indicates effectiveness of B over A.

If the material was effective in killing the flies, it is entirely possible that many of those flies that would have infested the walnuts in the check row were killed as a result of feeding during migratory stays on the treated foliage. The results indicated are of doubtful significance.

*Plot Experiment II.*—The data indicate that basic lead arsenate was more effective applied as a spray than as a dust (fig. 71). Approximately equal amounts of the actual toxic material should have been

Fig. 71.—Dusting equipment in operation in experimental control plots. (Photo taken at sunrise.)

retained by the tree in both plots. This result is contrary to results obtained in the laboratory toxicity studies.

Results indicate that a high degree of control was obtained in the spray plot.

*Plot Experiment III.*—This experiment was outlined to compare the efficacy of two and three applications of basic lead arsenate, with 10 and with 20 gallons per tree at each application. Results indicate that a high degree of control was obtained in all treated plots. The degree of infestation was too light in the treated plots to warrant comparison of the different treatments.

*Plot Experiment IV.*—The trees in this grove were relatively small and were interplanted with mature peach trees that were untreated. Results indicate that everything must be treated within a solid block to secure best results. Flies may reach egg-laying maturity on untreated

foliage, then migrate to treated trees and oviposit despite the presence of toxic materials.

*Plot Experiment V.*—This experiment was designed to determine whether or not the presence of sweetened material in the spray mixture increased the degree of control. The data indicate that it actually decreased the effectiveness of the lead arsenate, which is highly improbable. Flies were observed to feed freely on the spray material for several days after it was applied. This suggests variables that have not been accounted for. Slight foliage injury resulted where the syrup was used.

*Plot Experiment VI.*—This experiment was conducted to determine the merits of the bait spray as compared with full coverage of lead arsenate. The formula was similar to that used successfully in the control of the Mediterranean fruit fly with the exception of the substitution of basic lead arsenate for the acid lead arsenate. The sweetened material caused moderate foliage injury. Results indicate that the bait spray was inferior to the regular lead arsenate spray. The control in both plots was unsatisfactory.

*Plot Experiment VII.*—This was another test of the bait spray. Conclusions are unwarranted, because of the light infestation in both treated and check plots.

*Plot Experiment VIII.*—This experiment was designed to determine whether or not a higher degree of control could be obtained by increasing the dosage of lead arsenate, and also whether the addition of sweetened material resulted in still better control. The trees were of such size that 15 gallons of spray afforded satisfactory coverage. Results indicate superior performance of the lead arsenate where the sweetened material was omitted. This is apparently another instance of variables that are unaccounted for. Moderate foliage injury resulted from the use of the corn syrup.

*Plot Experiment IX.*—Data from this experiment indicate that 6 pounds of lead arsenate plus sweetened material, at 3 gallons per tree, is superior to 12 pounds of lead arsenate at 6 gallons per tree. This evidence is contradictory to that for experiments V, VI, and VIII, which show decreased control wherever sweetened material was used with lead arsenate; compare experiment VI B, which has the same formula as that used in IX B except that the former included cane sugar instead of beet sugar. In VI B the infestation was 13.9 per cent, while in IX B it was 3.6 per cent. Five gallons per tree was used in VI B, and 3 gallons per tree in IX B. Such erratic results are difficult to explain.

*Plot Experiment X.*—The dosage of basic lead arsenate was increased to 16 pounds to 100 gallons in this bait spray test. While a check was not maintained, the data indicate a high degree of control.

*Plot Experiment XI.*—The same material was used in plot A of this test as in experiment X with considerably poorer results. Straight basic lead arsenate dust produced a higher degree of control than did the bait spray, which contained 1.6 pounds of basic lead arsenate to 100 gallons. The data in this experiment appear significant; however the row of check trees unavoidably received more irrigation water than the remainder of the grove. This fact may have rendered the walnuts in this row more susceptible to oviposition.

*Discussion of Field Control Experiments in 1929.*—The generally erratic results obtained in the 1929 experiments afford very little reliable information; therefore only tentative conclusions may be drawn. However the following comments are justified: The degree of control obtained through the use of basic lead arsenate has been generally unsatisfactory. The addition of sweetened materials to the regular lead arsenate sprays did not increase their effectiveness. Bait sprays were no more promising than regular spray or dust applications of basic lead arsenate.

Plots of sufficient size and proper isolation are important features in obtaining significant data. Adequate check plots are indispensable in experiments of this nature. They can be dispensed with only when a reliable standard treatment has been developed, thereby enabling direct comparisons with other plots that are differentially treated.

The relation of irrigation practices to husk hardness at the time of maximum fly activity is not entirely understood. Since husk hardness appears quite definitely to be the most important factor governing susceptibility to infestation, it may have been partially responsible for the erratic results in these control studies.

*The Use of Sweet Materials in Control Practices.*—The success of bait sprays in Oregon in controlling the white-banded cherry fruit fly, *Rhagoletis cingulata* (Loew),[25] and also the use of sugar in population studies of *R. completa,* warranted investigation of the role of sweet materials. Chemotropic studies previously presented indicate that sucrose does not attract the walnut husk fly; however, when applied to the foliage, it does serve to congregate the flies; in moving about over the tree in search of food, they find the treated foliage and remain to engorge themselves. Fermenting New Orleans molasses offers some attraction. In population studies, 10 per cent sucrose solution sprayed onto small areas of foliage served to congregate the flies for only a few days. Therefore, tests were conducted to determine the length of time after application that sucrose and New Orleans molasses remain palatable to the fly.

TABLE 28

LENGTH OF TIME AFTER APPLICATION THAT SUCROSE AND NEW ORLEANS MOLASSES REMAIN PALATABLE*

| Concentration, per cent | Days after application | *R. completa* observed feeding | | | Drosophilids observed feeding | | | Visible evidence of material on foliage | | | Injury to foliage | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | Many | Few | None | Many | Few | None | Plain | Faint | None | Severe | Moderate | Slight | None |
| Granulated cane sugar spray | | | | | | | | | | | | | | |
| 5 | 1 | x | .... | .... | x | .... | .... | x | .... | .... | .... | .... | .... | x |
| | 2–3 | .... | x | .... | x | .... | .... | .... | x | .... | .... | .... | .... | x |
| | 4 | .... | .... | x | .... | x | .... | .... | .... | x | .... | .... | .... | x |
| | 5–6 | .... | .... | x | .... | .... | x | .... | .... | x | .... | .... | .... | x |
| 10 | 1 | x | .... | .... | x | .... | .... | x | .... | .... | .... | .... | .... | x |
| | 2 | .... | x | .... | x | .... | .... | x | .... | .... | .... | .... | .... | x |
| | 3 | .... | x | .... | x | .... | .... | .... | x | .... | .... | .... | .... | x |
| | 4 | .... | .... | x | .... | x | .... | .... | .... | x | .... | .... | .... | x |
| | 5–6 | .... | .... | x | .... | .... | x | .... | .... | x | .... | .... | .... | x |
| 25 | 1–3 | x | .... | .... | x | .... | .... | x | .... | .... | .... | .... | .... | x |
| | 4 | .... | x | .... | x | .... | .... | .... | x | .... | .... | .... | .... | x |
| | 5 | .... | .... | x | .... | x | .... | .... | x | .... | .... | .... | x | .... |
| | 6 | .... | .... | x | .... | x | .... | .... | x | .... | .... | x | .... | .... |
| 50 | 1–2 | x | .... | .... | x | .... | .... | x | .... | .... | .... | .... | .... | x |
| | 3 | x | .... | .... | x | .... | .... | x | .... | .... | .... | .... | x | .... |
| | 4 | x | .... | .... | x | .... | .... | x | .... | .... | .... | x | .... | .... |
| | 5 | .... | x | .... | x | .... | .... | x | .... | .... | .... | x | .... | .... |
| | 6 | .... | .... | x | .... | x | .... | .... | x | .... | x | .... | .... | .... |

* Observations extended from September 5 to 15, 1929.

TABLE 28—(*Concluded*)

| Concentration, per cent | Days after application | *R. completa* observed feeding | | | Drosophilids observed feeding | | | Visible evidence of material on foliage | | | Injury to foliage | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | Many | Few | None | Many | Few | None | Plain | Faint | None | Severe | Moderate | Slight | None |
| **Powdered cane sugar dust** | | | | | | | | | | | | | | |
| 100 | 1 | x | .... | .... | x | .... | .... | x | .... | .... | .... | .... | .... | x |
| | 2 | x | .... | .... | x | .... | .... | .... | x | .... | .... | .... | .... | x |
| | 3 | x | .... | .... | x | .... | .... | .... | .... | x | .... | .... | .... | x |
| | 4 | .... | x | .... | x | .... | .... | .... | .... | x | .... | .... | .... | x |
| | 5 | .... | .... | x | .... | x | .... | .... | .... | x | .... | .... | x | .... |
| | 6 | .... | .... | x | .... | x | .... | .... | .... | x | .... | x | .... | .... |
| **New Orleans molasses spray** | | | | | | | | | | | | | | |
| 10 | 1 | .... | x | .... | .... | x | .... | x | .... | .... | .... | .... | .... | x |
| | 2 | .... | x | .... | .... | x | .... | x | .... | .... | .... | .... | x | .... |
| | 3–6 | .... | .... | x | .... | x | .... | .... | x | .... | .... | .... | x | .... |
| 25 | 1 | .... | x | .... | .... | x | .... | x | .... | .... | .... | .... | .... | x |
| | 2 | x | .... | .... | x | .... | .... | x | .... | .... | .... | .... | x | .... |
| | 3–6 | .... | .... | x | .... | x | .... | .... | x | .... | .... | x | .... | .... |
| 50 | 1 | .... | x | .... | .... | x | .... | x | .... | .... | .... | .... | .... | x |
| | 2 | x | .... | .... | x | .... | .... | x | .... | .... | .... | x | .... | .... |
| | 3 | .... | x | .... | x | .... | .... | x | .... | .... | .... | x | .... | .... |
| | 4–6 | .... | .... | x | .... | x | .... | .... | x | .... | x | .... | .... | .... |

Tests made were in the same grove and flies were present in large numbers. Two small areas of foliage were treated on each of two trees in each individual test. The treated areas were observed for the presence of flies at an optimum time twice daily. The data obtained from these tests are presented in table 28.

After a period of eight days had elapsed, the applications were repeated on areas contiguous to those formerly treated. The later observations collaborated those previously recorded, lending significance to the data.

With cane sugar, the period over which the material remains palatable to the flies was in direct proportion to the concentration used. At concentrations of 5 and 10 per cent, this period is of 2 or 3 days' duration. No foliage injury was evident in the span of these tests from these concentrations of sugar. However, tree-tolerance tests have shown that 3 to 5 per cent concentrations of sucrose do cause slight injury. The indications are that powdered cane sugar applied as dust offers more promise in studies of this nature than granulated cane sugar in solution at any concentration.

New Orleans molasses is apparently not particularly palatable to the flies until about 24 hours after it has been applied. Furthermore, it is effective for about 24 hours only. At 10 per cent concentration, only a few flies were observed feeding upon it; while at 25 and 50 per cent concentrations many flies were congregated. The injury factor incident to its use is important.

In most instances drosophilids were observed feeding on the treated foliage for several days after it had lost its effectiveness in serving to congregate walnut husk flies.

On the basis of these limited tests, the addition of cane sugar or molasses to sprays or dusts of basic lead arsenate is of questionable value in the control of *Rhagoletis completa*. Furthermore bait sprays consisting of either of these sweet materials and a more rapidly acting poison than basic lead arsenate, do not appear very promising. Semiweekly applications during the period of adult emergence would probably be necessary in order to effect a satisfactory degree of control.

*Tree-Tolerance Studies.*—The effect of various materials upon walnut trees was determined in conjunction with laboratory toxicity studies of the same materials on the flies. These tests were made on seven-year-old Eureka walnuts at the Citrus Experiment Station. Because of the relatively large number of tests planned and the desirability of maintaining buffer trees between treated plots, it was necessary to restrict the size of plots to one tree. However, only normal trees were included in the tests. Most materials were applied both as spray and dust, one, two, and

TABLE 29

TOLERANCE OF EUREKA WALNUT TREES TO CERTAIN INSECTICIDAL MATERIALS*

| Material | Solubility in water at 20° C† | Concentration | Application | | Degree of injury§ | Remarks |
|---|---|---|---|---|---|---|
| | | | Method‡ | Number | | |
| Magnesium fluosilicate | 1 : 2.2 | 4 lbs.–100 gallons | Spray | 1<br>2<br>3 | Moderate<br>Severe<br>Severe | |
| | 1 : 2.2 | 33 per cent + talc | Dust | 1 | Slight | |
| Sodium fluoride | 1 : 25 | 33 per cent + talc | Dust | 1<br>2<br>3 | Moderate<br>Severe<br>Severe | |
| Sodium fluosilicate 98–100 per cent | 1 : 151 | 4 lbs.–100 gallons | Spray | 1<br>2<br>3 | Slight<br>Moderate<br>Severe | |
| | | 100 per cent (straight) | Dust | 1<br>2<br>3 | Slight<br>Moderate<br>Moderate | New growth shows first evidence of injury from fluorine compounds. Tips and margins of leaves, and also irregular areas between veins, have water-soaked appearance that later results in a "burned" appearance. Leaves of moderately to severely injured new growth usually drop, sections of twigs being defoliated, with new growth at the tips coming out after the treatment. |
| | | 33 per cent + talc | Dust | 1<br>2<br>3 | Very slight<br>Slight<br>Moderate | |
| Sodium fluosilicate 70–75 per cent | 1 : 151 | 4 lbs.–100 gallons | Spray | 1<br>2<br>3 | Very slight<br>Moderate<br>Severe | All fluorines tested exhibited relatively poor sticking qualities. However, sodium fluoaluminate and potassium fluoaluminate apparently adhered better than any of the other fluorine compounds. |
| | | 100 per cent (straight) | Dust | 1<br>2<br>3 | None<br>Very slight<br>Moderate | |
| Sodium fluoaluminate‖ (synthetic) | 1 : 1,639 | 4 lbs.–100 gallons | Spray | 1<br>2<br>3 | Very slight<br>Very slight<br>Slight | |
| | | 33 per cent + talc | Dust | 1<br>2<br>3 | None<br>Very slight<br>Slight | |
| | | 100 per cent (straight) | Dust | 1 | Slight | |

See page 528 for footnotes for this table.

TABLE 29—(*Continued*)

| Material | Solubility in water at 20° C† | Concentration | Application | | Degree of injury§ | Remarks |
|---|---|---|---|---|---|---|
| | | | Method‡ | Number | | |
| Potassium fluoaluminate¶ | 1 : 633 | 20 per cent + talc | Dust | 2 | None | See "Remarks," preceding page |
| Barium fluoride | 1 : 632 | 4 lbs.–100 gallons | Spray | 1<br>2<br>3 | None<br>None<br>Very slight | |
| | | 33 per cent + talc | Dust | 1<br>2<br>3 | None<br>None<br>Very slight | |
| Barium fluosilicate | 1 : 4,000 | 4 lbs.–100 gallons | Spray | 1<br>2<br>3 | None<br>Very slight<br>Slight | |
| | | 33 per cent + talc | Dust | 1<br>2<br>3 | None<br>None<br>Very slight | |
| | | 100 per cent (straight) | Dust | 1 | Slight | |
| Potassium fluosilicate | 1 : 1,130 | 33 per cent + talc | Dust | 1<br>2 | None<br>Very slight | |
| Calcium fluosilicate compound | 1 : 25,636 | 4 lbs.–100 gallons | Spray | 1<br>2<br>3 | None<br>None<br>Very slight | |
| | | 100 per cent (straight) | Dust | 1<br>2<br>3 | None<br>None<br>Very slight | |
| Calcium fluoride | 1 : 57,200 | 4 lbs.–100 gallons | Spray | 1<br>2<br>3 | None<br>None<br>Very slight | |
| | | 33 per cent + talc | Dust | 1<br>2<br>3 | None<br>None<br>Very slight | |

See page 528 for footnotes for this table.

TABLE 29—(*Continued*)

| Material | Solubility in water at 20° C† | Concentration | Application | | Degree of injury§ | Remarks |
|---|---|---|---|---|---|---|
| | | | Method‡ | Number | | |
| Sodium arsenite | Very soluble | 2 quarts–100 gallons | Spray | 1 | Very severe | Total defoliation and defruiting of trees. |
| | | 1 quart–100 gallons | Spray | 1 | Very severe | |
| Calcium arsenate | 1 : 133 | 2 lbs.–100 gallons | Spray | 1 | Slight | Typical arsenical injury is manifested in irregular burned areas between veins and along midrib of new and mature foliage. Moderately to severely injured leaves drop. Yellowing of leaves partially or totally, resulting in drop, also appears to be related to arsenical injury. |
| | | | | 2 | Moderate | |
| | | | | 3 | Severe | |
| | | 33 per cent + lime | Dust | 1 | Slight | |
| Acid lead arsenate | 1 : 200 | 3 lbs.–100 gallons | Spray | 1 | Moderate | |
| | | | | 2 | Severe | |
| | | | | 3 | Severe | |
| | | 100 per cent (straight) | Dust | 1 | Slight | |
| | | | | 2 | Severe | |
| | | | | 3 | Severe | |
| Basic lead arsenate | 1 : 200 | 4 lbs.–100 gallons | Spray | 1 | None | |
| | | | | 2 | None | |
| | | | | 3 | None | |
| | | 6 lbs.–100 gallons | Spray | 1 | None | |
| | | 12 lbs.–100 gallons | Spray | 1 | None | |
| | | 100 per cent (straight) | Dust | 1 | None | |
| | | | | 2 | None | |
| | | | | 3 | Very slight | |
| Magnesium arsenate¶ | 1 : 200 | 3 lbs.–100 gallons | Spray | 1 | None | |
| | | 20 per cent + lime | Dust | 2 | None | |
| Manganese arsenate¶ | 1 : 200 | 3 lbs.–100 gallons | Spray | 1 | Slight | |
| | | 20 per cent + lime | Dust | 1 | Slight | |
| Copper sulfate‖ | 1 : 5 | 4 lbs.–100 gallons | Spray | 1 | Moderate | Copper injury is manifested in moderate to large water-soaked areas between the veins occurring on both new and mature foliage. Affected leaves drop. Burn of margin and tip of leaf common but negligible in consequence. |
| | | | | 2 | Severe | |
| | | | | 3 | Very severe | |
| | | 10 per cent + lime | Dust | 1 | Slight | |
| | | | | 2 | Moderate | |
| | | 20 per cent + lime | Dust | 1 | Slight | |
| | | | | 2 | Moderate | |

See page 528 for footnotes for this table.

TABLE 29—(*Concluded*)

| Material | Solubility in water at 20° C† | Concentration | Application | | Degree of injury§ | Remarks |
|---|---|---|---|---|---|---|
| | | | Method‡ | Number | | |
| Copper carbonate‖ | Insoluble | 4 lbs.–100 gallons | Spray | 1<br>2<br>3 | Slight<br>Severe<br>Severe | See "Remarks" opposite "copper sulfate" on preceding page. |
| | | 20 per cent + lime | Dust | 1<br>2<br>3 | Slight<br>Slight<br>Moderate | |
| | | 90 per cent + lime | Dust | 1 | Severe | |
| Cane sugar (powdered) | | 100 per cent (straight) | Dust | 1 | Moderate | Typical injury from sweetened materials is manifested in a burning of leaf tips and margins, also large, irregular areas between veins. Leaves commonly turn yellow and drop without showing much evidence of being burned. |
| Beet sugar | | 25 lbs.–100 gallons | Spray | 1 | Slight | |
| "Black Strap Molasses" | | 40 lbs.–100 gallons | Spray | 1 | Slight | |
| Beet sugar<br>"Black Strap Molasses" | | 25 lbs.<br>40 lbs. –100 gallons | Spray | 1 | Moderate | |
| Basic lead arsenate<br>Beet sugar<br>"Black Strap Molasses" | | 6 lbs.<br>25 lbs.<br>40 lbs. –100 gallons | Spray | 1 | Moderate | |
| Basic lead arsenate<br>Beet sugar | | 6 lbs.<br>25 lbs. –100 gallons | Spray | 1 | Slight | |
| Basic lead arsenate<br>"Black Strap Molasses" | | 6 lbs.<br>40 lbs. –100 gallons | Spray | 1 | Slight | |

* All tests were in 1929 except where otherwise indicated.

† Solubilities of most fluorine compounds tested were determined by the Division of Chemistry, California State Department of Agriculture.

‡ Application was thorough in every instance. Subsequent applications were made at 2-week intervals.

§ All plots were carefully inspected weekly to determine effect of material on trees. Degree of injury is designated as follows:
Very slight—Trace of leaf injury.
Slight—Burn of tips and margins of leaves quite general over entire tree.
Moderate—Approximately one-fourth of total leaf area killed
Severe—Approximately one-half of total leaf area killed.
Very severe—Practically total defoliation.

‖ Tests conducted in 1930.

¶ Tests conducted in 1932.

three applications being made. Data obtained from these tests in 1929, together with data from tests of several other materials in 1930 and 1932, are presented in table 29.

The more soluble fluorine compounds were so injurious that they were eliminated from further trials. Considering tree tolerance, availability, and existing information regarding toxicity, barium fluosilicate and synthetic cryolite are more promising than any other fluorine compounds tested.

Tests of arsenicals corroborated conclusions of other workers in that basic lead arsenate is the only one of the commonly used arsenical compounds that can be applied with safety to walnut trees. However on the basis of one season's (1932) tests, magnesium arsenate appears promising with respect to safety to walnut foliage.

Copper sulfate and copper carbonate caused slight to severe injury at the concentrations tested. This is not entirely in accord with observations of phytopathologists.

The sweet materials tested resulted in slight to moderate injury. Studies previously reported indicate the questionable value of these materials when included in insecticide mixtures at the concentrations tested. No more injury was produced by the combination of sweet with basic lead arsenate than by the sweet material alone.

## FIELD INVESTIGATIONS IN 1930

In view of the somewhat erratic results obtained in 1929, an effort was made in 1930 to isolate all plots from each other and from surrounding vegetation. Furthermore this was desirable particularly where materials were applied as dust, since there is a certain amount of drift into adjoining trees, even under ideal conditions for application. Consequently four rows of trees surrounding each plot were sprayed with basic lead arsenate. This was considered standard treatment. These treated rows are hereafter referred to as "contact" areas. Figure 72 illustrates the typical method used in isolating plots (though these particular plots were not used in 1930).

More detailed attention was given to the fly population in the various plots in 1930. The basis of rating fly population numerically was not entirely satisfactory; however, it served as an index, at least, of the relative numbers of flies in various plots. The procedure was as follows:

When emergence records indicated that 50 to 75 per cent of the seasonal emergence had occurred, the rating of plots was begun. A 10 to 20 per cent solution of cane sugar was sprayed onto a small area on the northeast portion of a representative number of trees in each plot. The treated areas were in a position such that all flies present could be

readily observed. The time of observation with respect to temperature, humidity, and sunlight was comparable and, therefore, the records appear to be significant.

The arbitrary classes of 1, 2, 3, and 3+ were established to indicate the population found to exist as a result of these field studies. When the mean number of flies observed per tree was one, the rating of 1 was given; when from two to three, the rating of 2; when from four to five, the rating of 3; and when more than five, the rating of 3+.

There were no heavily infested groves in 1929, so that the experimental plots did not have a potentially high fly population from the annual generation. However, all plots were moderately to heavily infested in 1928. Since emergence in 1929 was only approximately 45 per cent of the total, the population potential of the biennial generations was relatively high.

Data regarding infestation in these control studies were based on field counts just prior to harvest. The "count trees" were located centrally within plots. Individual tree records were kept to enable statistical treatment of the data. For calculating probable error of the mean, the Gaussian formula was used in those instances where the number of count trees was 16 or more. Since in most tests the number of count trees was under 16, the Bessel formula was used for most of the calculations. These standard formulas are as follows:[20]

$$\text{Gaussian: P.E.} = \pm 0.6745 \times \frac{\sigma}{\sqrt{N}}$$

$$\text{Bessel: P.E.} = \pm \frac{0.6745 \times \sigma}{\sqrt{N-1}}$$

Adult emergence began in the middle of July and the seasonal peak was reached shortly thereafter on July 18 (fig. 39). Plots were differentially treated; however, in all instances one application was made before the peak was reached. The data obtained from these field control experiments are presented in table 30.

*Plot Experiment I.*—This experiment was outlined to determine the efficacy of one application of basic lead arsenate spray as compared with that of two applications. Since differences in percentage of infestation between untreated plots varied as greatly as those between treated and untreated plots, important conclusions are unwarranted. However, the percentages of infestation in plots B and C indicate a superiority of two spray applications over one application.

*Plot Experiment II.*—It was desirable to determine the relative merits of basic lead arsenate applied as spray and as dust; also the effect of a single application just prior to the peak of adult emergence. Concerning the latter, the application in plot B was made on August 12, in the

TABLE 30

CONTROL EXPERIMENTS IN 1930

| Experiment | Number of trees and variety | Material | Concentration | Application: Method | Application: Amount per tree | Application: Number | Application: Date | Estimated per cent infested 1929* | Population rating† | Results: Trees counted | Results: Total nuts counted | Results: Mean per cent infested‡ |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| I | (10 acres) | | | | | | | | | | | |
| *A* | 25 Eureka | Basic lead arsenate | 4 lbs.–100 gals. | Spray | 15 gals. | 1 | 7/17 | 1–2 | 1 | 4 | 2,104 | 2.2±0.6 |
| *B* | 24 Eureka | Basic lead arsenate | 4 lbs.–100 gals. | Spray | 15 gals. | 2 | 7/17, 8/3 | 1–2 | 2 | 4 | 2,240 | 0.9±0.6 |
| *C* | 92 Eureka§ | Basic lead arsenate | 4 lbs.–100 gals. | Spray | 15 gals. | 2 | 7/17, 8/3 | 1–2 | 2 | 4 | 2,342 | 1.0±0.6 |
| *D* | 24 Eureka | Check, no treatment | ........ | ........ | ........ | .... | ........ | 1–2 | 2 | 4 | 1,933 | 2.0±0.5 |
| *E* | 24 Eureka | Check, no treatment | ........ | ........ | ........ | .... | ........ | 1–2 | 2 | 4 | 2,193 | 6.2±1.8 |
| *D-E* | ........ | ........ | ........ | ........ | ........ | .... | ........ | 1–2 | 2 | 8 | 4,126 | 4.2±1.1 |
| II | (10 acres) | | | | | | | | | | | |
| *A* | 32 Eureka | Basic lead arsenate | 4 lbs.–100 gals. | Spray | 15 gals. | 1 | 7/16 | 4–6 | 2 | 4 | 2,141 | 6.2±1.0 |
| *B* | 32 Eureka | Basic lead arsenate | 4 lbs.–100 gals. | Spray | 15 gals. | 1 | 8/12 | 4–6 | 1 | 4 | 2,160 | 3.8±0.8 |
| *C* | 32 Eureka | {Basic lead arsenate<br>{Hydrated lime | 20 lbs.}<br>80 lbs.} | Dust | 3 lbs. | 1 | 7/16 | 4–6 | 2 | 4 | 2,152 | 12.2±3.7 |
| *D* | 32 Eureka | {Basic lead arsenate<br>{Hydrated lime | 20 lbs.}<br>80 lbs.} | Dust | 3 lbs. | 2 | 7/16, 8/13 | 4–6 | 3 | 4 | 2,209 | 6.4±1.4 |
| *E* | 95 Eureka§ | Basic lead arsenate | 4 lbs.–100 gals. | Spray | 15 gals. | 2 | 7/16, 8/13 | 4–6 | 2 | 4 | 1,721 | 2.5±0.6 |
| III | (3½ acres) | | | | | | | | | | | |
| *A* | 12 Eureka + 42 peach | Walnuts: Basic lead arsenate | 4 lbs.–100 gals. | Spray | 15 gals. | 3 | 7/16, 8/3, 8/20 | 4–6 | 3 | 4 | 2,446 | 11.5±1.2 |
| *A1* | 20 Eureka + 51 peach | Peaches: {Basic lead arsenate<br>Powdered cane sugar | 9 lbs.<br>1 lb. | Dust | 1½ oz. | 2 | 7/18, 8/5 | 4–6 | 2 | 4 | 1,533 | 11.1±1.4 |
| *A-A1* | ........ | ........ | ........ | ........ | ........ | .... | ........ | ..... | 3 | 8 | 3,979 | 11.3±0.7 |
| *B* | 12 Eureka + 42 peach | Walnuts: Basic lead arsenate | 4 lbs.–100 gals. | Spray | 15 gals. | 3 | 7/16, 8/3, 8/20 | 4–6 | 3 | 4 | 1,431 | 11.7±2.6 |
| *B1* | 18 Eureka + 48 peach | Peaches: check, no treatment | ........ | ........ | ........ | .... | ........ | 4–6 | 3 | 4 | 2,010 | 17 8±0.9 |
| *B-B1* | ........ | ........ | ........ | ........ | ........ | .... | ........ | 4–6 | 3 | 8 | 3,441 | 15.2±1.6 |

* All plots were moderately to heavily infested in 1928; thus the biennial brood would greatly augment the fly population in 1930.

† See pp. 529-530 for method of determining population rating.

‡ Data based on count of nuts on trees just prior to harvest (October 10); percentage infested with walnut husk fly except where otherwise indicated.

§ These were trees surrounding experimental plots for purpose of isolation, called "contact trees" in the text.

TABLE 30—(*Concluded*)

| Experiment | Number of trees and variety | Material | Concentration | Application: Method | Application: Amount per tree | Application: Number | Application: Date | Estimated per cent infested 1929* | Population rating† | Results: Trees counted | Results: Total nuts counted | Results: Mean per cent infested‡ |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| IV | (5 acres)¶ | | | | | | | | | | | |
| *A* | 85 Eureka | Basic lead arsenate | 4 lbs.–100 gals. | Spray | 10 gals. | 1 | 7/17 | 4–6 | 1 | 16 | 2,116 | 3.9±0.3 |
| *A1* | 20 Eureka | Basic lead arsenate<br>Powdered cane sugar | 9 lbs.<br>1 lb. | Dust | 3 oz. | 2 | 7/18, 8/17 | 4–6 | 1 | 7 | 1,178 | 5.3±0.6 |
| *A2* | 26 Eureka | Check, no treatment | ........ | ........ | ........ | ........ | ........ | 4–6 | 2 | 15 | 1,422 | 2.3±0.4 |
| *B* | 35 Payne | Basic lead arsenate<br>Powdered cane sugar | 9 lbs.<br>1 lb. | Dust | 3 oz. | 2 | 7/18, 8/17 | 2–4 | 2 | 14 | 1,751 | 0.0 |
| *B1* | 16 Payne | Check, no treatment | ........ | ........ | ........ | ........ | ........ | 2–4 | 1 | 16 | 1,782 | 0.1±0.0 |
| V | (12 acres)‖ | | | | | | | | | | | |
| *A* | 24 walnut | Basic lead arsenate | 4 lbs.–100 gals. | Spray | 5 gals. | 2 | 7/17, 8/12 | 10–30 | 2 | ....** | ........ | 15.5†† |
| *B* | 24 walnut | Basic lead arsenate | 4 lbs.–100 gals. | Spray | 10 gals. | 2 | 7/17, 8/12 | 10–30 | 1 | .... | ........ | 35.0†† |
| *C* | 48 walnut | Barium fluosilicate††<br>Talc | 20 lbs.<br>80 lbs. | Dust | 2 lbs. | 2 | 7/16, 8/14 | 10–30 | 1 | .... | ........ | 3.4†† |
| *D* | 128 walnut§ | Basic lead arsenate | 4 lbs.–100 gals. | Spray | 15 gals. | 2 | 7/17, 8/12 | 10–30 | 1 | .... | ........ | ........ |
| *E* | 24 walnut | Check, no treatment | ........ | ........ | ........ | .... | ........ | 10–30 | 2 | .... | ........ | 12.8†† |
| VI | (10 acres) | | | | | | | | | | | |
| *A* | 42 Eureka | Barium fluosilicate‡‡<br>Talc | 20 lbs.<br>80 lbs. | Dust | 2 lbs. | 2 | 7/18, 8/13 | 1–2 | 1 | 10 | 4,154 | 0.9±0.2 |
| *B* | 10 Eureka | Basic lead arsenate<br>Hydrated lime | 20 lbs.<br>80 lbs. | Dust | 2 lbs. | 2 | 7/18, 8/13 | 2–4 | 1 | 2 | 449 | 36.7±6.6 |
| *C* | 70 Eureka§ | Basic lead arsenate | 4 lbs.–100 gals. | Spray | 15 gals. | 2 | 7/16, 8/12 | 1–2 | 1 | .... | ........ | ........ |
| *D* | 42 Eureka | Check, no treatment | ........ | ........ | ........ | .... | ........ | 1–2 | 1 | 9 | 2,621 | 8.3±1.9 |
| Totals, 27 plots, 1,068 walnut trees, 183 peach trees, 51 acres | | | | | ........ | ........ | ........ | .... | .... | 145 | 44,088 | ........ |

* All plots were moderately to heavily infested in 1928; thus the biennial brood would greatly augment the fly population in 1930.

† See pp. 529-530 for method of determining population rating.

‡ Data based on count of nuts on trees just prior to harvest (October 10); percentage infested with walnut husk fly except where otherwise indicated.

§ These were trees surrounding experimental plots for purpose of isolation, called "contact trees" in the text.

¶ Small trees.

‖ Assorted varieties of large trees; grove proved unadapted for desired experiments

** Not practicable to make counts since trees are very old and tall, and pruned high.

†† Percentage of harvest culls, including nuts injured by walnut husk fly, blight, sunburn, and codling moth.

‡‡ Slight foliage injury, though negligible.

belief that the seasonal peak of emergence was yet to be reached, while actually more than 70 per cent of the total emergence had taken place by that date. The data indicate results superior to one application made just prior to the peak, and also superior to two applications of the material as dust. A possible explanation lies in the fact that the material was more readily available to a larger percentage of the total flies, since it was applied later and thus was not covered with dust from the soil as a result of orchard cultivation. This test shows that appreciable oviposition did not take place, at least within 30 days after the seasonal peak of emergence—a fact of interest and importance.

These tests demonstrate that under existing conditions basic lead arsenate is more efficacious when applied as spray than as dust.

*Plot Experiment III.*—This experiment was designed to determine the value of treating trees other than walnuts that are growing as interplants. All walnut trees were sprayed with basic lead arsenate at the regularly used concentration and coverage. Peach trees in half of the grove were treated with a poison bait, which was applied mainly to the nonbearing sucker growth in the centers of the trees. Peach trees in the remaining half of the grove were untreated and served as a check. The results indicate a slight reduction in the infestation where the peaches were treated. Considering the results obtained in other experiments with two spray applications of basic lead arsenate, it is evident that the material applied to the peach trees was of little value. Furthermore the indications are that the faliure to effect a higher degree of control on walnuts is directly attributable to the fact that the peaches did not receive effective treatments.

*Plot Experiment IV.*—This experiment was conducted to compare the efficacy of basic lead arsenate used in a "bait" dust, with that applied as spray with complete coverage. The results afford no information regarding control, but they do point out the nature of some of the uncontrollable variations—for instance, a higher degree of infestation occurred in the tested plots A and A1 than in the check A2, while a higher fly population existed in plot A2.

*Plot Experiment V.*—The trees in this grove were assorted varieties, which vary in susceptibility to attack. Besides, they were old, tall, and pruned high; and on the whole were not entirely satisfactory for experimental use. The purpose of the experiment was to determine the efficacy of a very light spray application of basic lead arsenate, and of barium fluosilicate applied as dust. The percentage of infestation could not be satisfactorily determined in the field; therefore the percentage of cull walnuts in each plot was obtained from the harvest record. While these data are not truly signficant with respect to the degree of infestation of

the walnut husk fly, they indicate roughly the relative effectiveness of the various treatments. Barium fluosilicate apparently effected a considerable reduction in the degree of infestation. This material caused slight foliage injury, particularly to the newest growth, although the injury was negligible in extent. A light application of lead arsenate is apparently of little value in controlling the fly.

*Plot Experiment VI.*—This experiment was a comparison of barium fluosilicate and basic lead arsenate applied as dust. As evidenced by the results obtained, barium fluosilicate is greatly superior to basic lead arsenate. Slight foliage injury resulted from the use of the former material; however, it was negligible in degree.

The data from this experiment illustrate how the degree of infestation may vary within a grove. For instance, the infestation in plot B, after two dust applications, was 36 per cent; while in plot D, which was not treated, the infestation was 8 per cent.

*Discussion of Field Control Experiments in 1930.*—The fact that in most instances the probable error of the mean percentage infested is less than one-third of the uncorrected datum, indicates significance of the data.

In summarizing the results obtained from the various control experiments, the following conclusions appear justifiable: Two spray applications of basic lead arsenate, 4 pounds to 100 gallons, offer a fairly satisfactory means of controlling the walnut husk fly. The efficacy of this material applied as a spray is superior to dust applications; in fact, one spray treatment is approximately equivalent to two dust treatments. Barium fluosilicate is sufficiently promising to warrant further investigation. For the best results, all interplanted trees and other vegetation within the walnut grove proper, together with that immediately bordering the outside rows, should be treated. Isolation of experimental plots is essential. The method employed for this purpose in these studies is satisfactory and practicable. Accurate comparisons of results obtained from the use of the same materials in different groves are not possible, because of differences in soil types, irrigation, and other cultural practices, as well as the size of the crop of nuts that is developing. When fly populations are comparable, the latter feature apparently bears some relation to percentage infestation.

The fact that in most instances the population rating of treated plots was practically the same as that of untreated plots suggests that the method of obtaining this index did not furnish detailed information with a high degree of accuracy. Therefore, its use is limited and conclusions based thereon require qualification. However, on the assumption that the method is reasonably reliable, there apparently is no signifi-

cant relation between the number of flies present in a plot and the resulting percentage of infested nuts. If true, it is logical to suspect that not all of the walnuts were in an optimum condition for females to puncture the husks in oviposition. In this connection, with reference to experiment I, notes on observations of the abundance of flies present in 1928 indicate that they were no more plentiful then than during the 1930 season. In 1928 the infestation was estimated to be between 70 and 90 per cent; while in 1930 it did not exceed 6 per cent in any of the plots.

## FIELD INVESTIGATIONS IN 1931

In the field plot trials for 1931 virtually the same procedure was followed as in the 1930 studies except that the contact areas were sprayed with barium fluosilicate instead of basic lead arsenate. However, in some respects more detailed information was desirable. This information regarding history of the plots in 1930 seemed necessary as an aid in planning experiments and laying out plots for the 1931 control studies. In many of the experiments actual tree counts of the number of infested nuts per tree in 1930 were made in order to obtain an idea of the homogeneity of the infestation. It was not practicable to obtain this information in all groves, nor was the percentage of infestation determined in some instances where counts were made of infested nuts per tree. Furthermore, in some plots the percentage of infestation was estimated. Where data are given for individual plots these are relative to other plots within the experiment.

A larger number of count trees were used in the 1931 experiments than in previous studies. Since control plots were maintained in most instances, the apparent percentage of reduction in infestation (or percentage of control), resulting from the use of the various materials, was determined. For these computations the commonly used formula discussed in the 1929 control studies was employed. However, as used in the 1931 studies $\overline{A}$ and $\overline{B}$ are both affected by probable errors. Therefore it was necessary to calculate the probable error of the quotient $\frac{\overline{B} \pm b}{\overline{A} \pm a}$. In these instances the following formula was used:[20]

$$Eq = \pm \frac{\sqrt{\left(\frac{\overline{B}a}{\overline{A}}\right)^2 + b^2}}{\overline{A}}$$

Adult emergence began during the first week of July and the seasonal peak was reached a few days afterward on July 12 (fig. 42). Thus emergence was earlier than in any of the preceding seasons of this study.

The data obtained from these field control plots are summarized in table 31.

TABLE 31

Control Experiments in 1931 *Columns continued on next page*

| Experiment | Number of trees and variety | Material | Concentration (per 100 gals. for sprays and per 100 lbs. for dusts) | Method of application | Amount per tree, gals. for sprays and lbs. for dusts |
|---|---|---|---|---|---|
| I | (44 acres)<br>Peach interplants | | | | |
| *A* | 48 Eureka + 48 peach | Barium fluosilicate<br>Talc | 20 lbs.<br>80 lbs. | Dust | W[c] 2<br>P[c] ½ |
| *B* | 48 Eureka + 48 peach | Barium fluosilicate<br>Diatomaceous earth<br>Fish oil[e] | 20 lbs.<br>80 lbs.<br>2 pts. | Dust | W 2<br>P ½ |
| *C* | 48 Eureka + 48 peach | Barium fluosilicate<br>Talc | 20 lbs.<br>80 lbs. | Dust | W 2<br>P ½ |
| *D* | 48 Eureka + 48 peach | Barium fluosilicate | 3 lbs. | Spray | W 15<br>P 5 |
| *E* | 48 Eureka + 48 peach (peaches not treated) | Barium fluosilicate | 3 lbs. | Spray | W 15<br>P 0 |
| *F* | 416 Eureka[f] + 416 peach[f] | Barium fluosilicate | 3 lbs. | Spray | W 15<br>P 5 |
| *G* | 48 Eureka + 48 peach | Check No. 1, no treatment | ........ | ........ | ........ |
| *H* | 48 Eureka + 48 peach | Check No. 2, no treatment | ........ | ........ | ........ |
| II | (7 acres)<br>Placentia interplants | | | | |
| *A* | 12 Eureka + 12 Placentia | Barium fluosilicate<br>Talc | 20 lbs.<br>80 lbs. | Dust | 3 |
| *B* | 12 Eureka + 12 Placentia | Barium fluosilicate<br>Talc | 20 lbs.<br>80 lbs. | Dust | 3 |
| *C* | 12 Eureka + 12 Placentia (Placentias not treated) | Barium fluosilicate<br>Talc | 20 lbs.<br>80 lbs. | Dust | 3 |
| | 12 Eureka + 12 Placentia (Placentias not treated) | Barium fluosilicate<br>Talc | 20 lbs.<br>80 lbs. | Dust | 3 |
| *E* | 18 Eureka + 12 Placentia | Check, no treatment | ........ | ........ | ........ |
| III | (5½ acres)<br>Small walnut trees, peach interplants | | | | |
| *A* | 34 Eureka + 112 peach | Barium fluosilicate | 3 lbs. | Spray | W 7<br>P 5 |
| *B* | 36 Eureka + 112 peach (peaches not treated) | Barium fluosilicate | 3 lbs. | Spray | W 7<br>P 0 |
| *C* | 24 Eureka + 72 peach | Check, no treatment | ........ | ........ | ........ |

c W=Walnut; P=peach.

e Fish oil was classified as "light-pressed."

f These were trees surrounding experimental plots for purpose of isolation, called "contact trees" in text.

TABLE 31—(*Continued*)

| Number of applications | Date | Foliage injury "burn" | Average infestation, 1930 | | Population rating[a] | Results[b] | | | Per cent control |
|---|---|---|---|---|---|---|---|---|---|
| | | | Nuts per tree | Per cent | | Trees counted | Total nuts counted | Mean per cent infested | |
| 1 | 7/9 | W Slight[d]<br>P None | 17 | .... | 2 | 28 | 4,182 | 16.9±1.0 | 45.0± 4.5 |
| 1 | 7/14 | W Slight<br>P None | 7 | .... | 1 | 27 | 8,189 | 10.3±2.2 | 66.5± 7.0 |
| 2 | 7/9, 8/17 | W Slight<br>P None | 5 | .... | 1 | 27 | 7,299 | 2.7±0.2 | 91.2± 4.5 |
| 1 | 7/14 | W Slight<br>P None | 3 | .... | 1 | 26 | 9,952 | 6.2±0.4 | 79.8± 4.5 |
| 1 | 7/14 | W Slight<br>P None | 4 | .... | 1 | 27 | 8,237 | 17.1±1.1 | 44.3± 4.5 |
| 1 | 7/18 | W Slight<br>P None | 3 | .... | 1 | 12 | 4,460 | 5.9±0.4 | 80.8± 4 5 |
| .... | .......... | .......... | 9 | .... | 2 | 28 | 6,924 | 30.7±1.8 | .......... |
| .... | .......... | .......... | 1 | .... | 1 | 28 | 10,238 | 6.9±0.8 | .......... |
| 1 | 7/15 | Slight | .... | 5[g] | 1 | 12 | 1,930 | 3.6±0.7 | 89.0± 4.3 |
| 2 | 7/15, 8/18 | Slight | | | 1 | 12 | 2,129 | 4.8±0.6 | 85.3± 4.3 |
| 1 | 7/15 | Slight | | | 1 | 12 | 1,744 | 8.4±0.7 | 74.3± 4.3 |
| 2 | 7/15, 8/18 | Slight | | | 1 | 12 | 2,235 | 3.2±0.5 | 90.2± 4.3 |
| .... | .......... | .......... | | | 3 | 18 | 2,970 | 32.6±2.4 | .......... |
| 1 | 7/10 | Slight | 19 | 65 | 1 | 33 | 1,128 | 2.2±0.4 | 90.4± 6.2 |
| 1 | 7/10 | Slight | 22 | 51 | 1 | 35 | 726 | 2.6±0.5 | 88.7± 6.2 |
| .... | .......... | .......... | 15 | 55 | 2 | 21 | 418 | 22.9±1.9 | .......... |

[a] See pp. 527-530 for method of determining population rating.
[b] Data based on count of nuts on trees just prior to harvest (September 28).
[d] "Slight"—injury is negligible.
[g] Estimated average for entire grove.

TABLE 31—*(Continued)*

*Columns continued on next page*

| Experiment | Number of trees and variety | Material | Concentration (per 100 gals. for sprays and per 100 lbs. for dusts) | Method of application | Amount per tree, gals. for sprays and lbs. for dusts |
|---|---|---|---|---|---|
| IV | (3½ acres) | | | | |
| *A* | 30 Eureka | Barium fluosilicate<br>Talc | 20 lbs.<br>80 lbs. | Dust | 3 |
| *B* | 30 Eureka | Basic lead arsenate<br>Hydrated lime | 20 lbs.<br>80 lbs. | Dust | 3 |
| V | (12 acres)<br>Placentia interplants | | | | |
| *A* | 40 Eureka + 40 Placentia | Barium fluosilicate<br>Fish oil[c] | 3 lbs.<br>1 pt. | Spray | 10 |
| *B* | 30 Eureka + 30 Placentia | Hydrated lime | 100 lbs. | Dust | 3 |
| *C* | 30 Eureka + 30 Placentia | Check, no treatment | ........ | ........ | ........ |
| VI | (2½ acres) | | | | |
| *A* | 3 Eureka (isolated in Placentia grove) | Barium fluosilicate | 3 lbs. | Spray | 15 |
| *B* | 4 Eureka + 32 Placentia (Eurekas isolated in Placentia grove—barrier of Placentias 1 tree deep treated surrounding individual trees) | Barium fluosilicate | 3 lbs. | Spray | 15 |
| *C* | 4 Eureka (isolated in Placentia grove) | Check, no treatment | ........ | ........ | ........ |
| VII | (2 acres) | | | | |
| *A* | 2 Eureka (isolated in Placentia grove) | Cryolite (synthetic) | 3 lbs. | Spray | 15 |
| *B* | 3 Eureka + 24 Placentia (Eurekas isolated in Placentia grove—barrier of Placentias 1 tree deep treated surrounding individual trees) | Cryolite (synthetic) | 3 lbs. | Spray | 15 |
| *C* | 3 Eureka (isolated in Placentia grove) | Check, no treatment | ........ | ........ | ........ |
| VIII | (2½ acres) | | | | |
| *A* | 3 Eureka (isolated in Placentia grove) | Cryolite (natural)<br>Diatomaceous earth<br>Fish oil[c] | 20 lbs<br>80 lbs.<br>2 lbs. | Dust | 2 |
| *B* | 4 Eureka + 32 Placentia (Eurekas isolated in Placentia grove—barrier of 1 tree deep treated surrounding individual trees) | Cryolite (natural)<br>Diatomaceous earth<br>Fish oil[c] | 20 lbs.<br>80 lbs.<br>2 lbs. | Dust | 2 |
| *C* | 4 Eureka (isolated in Placentia grove) | Check, no treatment | ........ | ........ | ........ |

[a] See pp. 529-530 for method of determining population rating.
[b] Data based on count of nuts on trees just prior to harvest (September 28).
[c] Fish oil was classified as "light-pressed."

TABLE 31—*(Continued)*

| Number of applications | Date | Foliage injury "burn" | Average infestation, 1930 | | Population rating[a] | Results[b] | | | Per cent control |
|---|---|---|---|---|---|---|---|---|---|
| | | | Nuts per tree | Per cent | | Trees counted | Total nuts counted | Mean per cent infested | |
| 1 | 7/10 | Slight | 65 | .... | 3 | 30 | 2,052 | 58.9±1.8 | 28.9± 3.4[h] |
| 1 | 7/10 | None | 29 | .... | 3 | 30 | 2,499 | 82.8±2.2 | .................... |
| 1 | 7/14 | Slight | | | 1 | 39 | 2,193 | 3.1±0.3 | 91.7± 3.8 |
| 1 | 7/13 | None | .... | 5[g] | 1 | 26 | 746 | 7.1±1.6 | 80.9± 5.9 |
| .... | ................ | .................... | | | 2 | 24 | 772 | 37.1±1.9 | .................... |
| 1 | 7/10 | Slight | | | 3 | 3 | 792 | 39.0±7.3 | 38.6±13.7 |
| 1 | 7/10 | Slight | .... | 50[i] | 2 | 4 | 1,432 | 26.5±6.8 | 58.3±12.9 |
| .... | ................ | .................... | | | 3 | 4 | 868 | 63.5±8.5 | .................... |
| 1 | 7/14 | Slight | | | 1 | 2 | 236 | 13.1±5.5 | 77.7±10.7 |
| 1 | 7/14 | Slight | .... | 40[i] | 1 | 3 | 179 | 31.2±6.2 | 46.8±13.3 |
| .... | ................ | .................... | | | 2 | 3 | 319 | 58.6±8.7 | .................... |
| 1 | 7/14 | Slight | | | 2 | 3 | 656 | 59.6±5.0 | 11.6±12.9 |
| 1 | 7/14 | Slight | .... | 50[i] | 2 | 4 | 1,296 | 68.1±4.7 | +1.0±14.1[j] |
| .... | ................ | .................... | | | 2 | 4 | 575 | 67.4±7.7 | .................... |

[g] Estimated average for entire grove.
[h] Indicates increased effectiveness of barium fluosilicate over basic lead arsenate.
[i] Estimated average for all Eureka trees in grove.
[j] Actual increase in degree of infestation over check.

TABLE 31—*(Continued)*

*Columns continued on next page*

| Experiment | Number of trees and variety | Material | Concentration (per 100 gals. for sprays and per 100 lbs. for dusts) | Method of application | Amount per tree, gals. for sprays and lbs. for dusts |
|---|---|---|---|---|---|
| IX | (10 acres) Neff interplants | | | | |
| *A* | 16 Eureka + 16 Neff | Cryolite (synthetic)<br>Talc | 20 lbs.<br>80 lbs. | Dust | 3 |
| *B* | 15 Eureka + 16 Neff | Cryolite (natural)<br>Talc | 20 lbs.<br>80 lbs. | Dust | 3 |
| *C* | 15 Eureka + 16 Neff | Talc | 100 lbs. | Dust | 3 |
| *D* | 42 Eureka[f] + 42 Neff[f] | Barium fluosilicate | 3 lbs. | Spray | 15 |
| *E* | 16 Eureka + 16 Neff | Check, no treatment | ...... | ...... | ...... |
| X | (2 acres) Franquette interplants | | | | |
| *A* | 6 Franquette + 12 Placentia | Cryolite (synthetic) | 100 lbs. | Dust | 2 |
| *B* | 4 Franquette + 10 Placentia | Check, no treatment | ...... | ...... | ...... |
| XI | (1½ acres) Eureka interplants | | | | |
| *A* | 4 Eureka + 16 Placentia | Nicotine sulfate 40%<br>Butanol<br>White oil 80 sec. visc. | 2½ pts.<br>2½ pts.<br>3 gals. | Spray | 15 |
| *B* | 4 Eureka | Check, no treatment | ...... | ...... | ...... |
| XII | (3 acres) | | | | |
| *A* | 20 Eureka | Nicotine sulfate 40%<br>Tannic acid | 1 pt.<br>3 lbs. | Spray | 15 |
| B | 15 Eureka | Barium fluosilicate | 3 lbs. | Spray | 15 |
| *C* | 12 Eureka | Check, no treatment | ...... | ...... | ...... |
| XIII | (3 acres) Eureka interplants | | | | |
| *A* | 12 Payne + 6 Eureka | Nicotine sulfate 40%<br>Sucrose | 1 pt.<br>12 lbs. | Spray | 15 |
| *B* | 10 Payne + 9 Eureka | Nicotine sulfate 40%<br>Bentonite | 5 lbs.<br>95 lbs. | Dust | 3 |
| *C* | 5 Payne + 5 Eureka | Check, no treatment | ...... | ...... | ...... |

[a] See pp. 529-530 for method of determining population rating.
[b] Data based on count of nuts on trees just prior to harvest (September 28).
[f] These were trees surrounding experimental plots for purpose of isolation, called "contact trees" in text.
[i]Estimated average for all Eureka trees in grove.

TABLE 31—*(Continued)*

| Number of applications | Date | Foliage injury "burn" | Average infestation, 1930 | | Population rating[a] | Results[b] | | | Per cent control |
|---|---|---|---|---|---|---|---|---|---|
| | | | Nuts per tree | Per cent | | Trees counted | Total nuts counted | Mean per cent infested | |
| 1 | 7/13 | Slight | 66 | 12 | 1 | 16 | 2,954 | 8.5±1.5 | 62.4± 9.7 |
| 2 | 7/13, 8/17 | Slight | 33 | 6 | 1 | 15 | 2,165 | 3.5±0.5 | 84.5± 6.2 |
| 2 | 7/13, 8/1 | None | 20 | 4 | 2 | 15 | 2,543 | 29.2±3.4 | +29.0±22.0[j] |
| 1 | 7/12 | Slight | 11 | 3 | 1 | 5 | 572 | 12.5±3.6 | 44.7±19.8 |
| .... | .......... | .......... | 36 | 6 | 2 | 16 | 2,513 | 22.6±3.2 | .......... |
| 1 | 7/15 | Slight | .... | 75[k] | 1 | 6 | 223 | 1.7±0.4 | .......... |
| .... | .......... | .......... | | | 1 | 4 | 257 | 0.0 | .......... |
| 2 | 7/15, 8/3 | Moderate | .... | 60[i] | 2 | 4 | 812 | 78.0±5.5 | +15.0±17.4[j] |
| .... | .......... | .......... | | | 3 | 4 | 575 | 67.4±8.5 | .......... |
| 2 | 7/13, 8/5 | None | .... | 30[i] | 1 | 10 | 2,586 | 13.2±1.8 | 74.4± 5.3 |
| 2 | 7/13, 8/5 | Slight | | | 1 | 8 | 2,082 | 8.2±2.6 | 84.1± 6.1 |
| .... | .......... | .......... | | | 3+ | 9 | 2,693 | 51.4±8.8 | .......... |
| 2 | 7/11, 8/2 | Slight | .... | 40[l] | 3 | 13 | 2,685 | 44.4±6.0 | 45.3± 8.0 |
| 2 | 7/12, 8/5 | None | | | 1 | 19 | 3,070 | 39.2±4.8 | 51.7± 6.9 |
| .... | .......... | .......... | | | 3 | 7 | 1,950 | 81.1±6.0 | .......... |

[j] Actual increase in degree of infestation over check.
[k] Estimated average for all Franquette trees in grove.
[l] Estimated average for all Payne and Eureka trees in grove.

TABLE 31—*(Concluded)*

*Columns continued on next page*

| Experiment | Number of trees and variety | Material | Concentration (per 100 gals. for sprays and per 100 lbs. for dusts) | Method of application | Amount per tree, gals. for sprays and lbs. for dusts |
|---|---|---|---|---|---|
| XIV | (5 acres) | | | | |
| *A* | 20 Eureka + 19 seedling | Nicotine sulfate 40%<br>Diatomaceous earth | 5 lbs.<br>95 lbs. | Dust | 2 |
| *B* | 20 Franquette | Barium fluosilicate<br>Talc | 20 lbs.<br>80 lbs. | Dust | 2 |
| *C* | 15 Franquette + 5 Eureka | Check, no treatment | ........ | ........ | ........ |
| XV | (10 acres) | | | | |
| *A* | 25 Eureka | Tobacco dust | 100 lbs.[t] | Dust | 2 |
| *B* | 25 Eureka | Diatomaceous earth | 100 lbs. | Dust | 2 |
| *C* | 25 Eureka | Barium fluosilicate | 100 lbs. | Dust | 1½ |
| *D* | 70 Eureka[f] | Barium fluosilicate | 3 lbs. | Spray | 20 |
| *E* | 25 Eureka | Check, no treatment | ........ | ........ | ........ |
| XVI | (1/3 acre) | | | | |
| *A* | 3 Klondike | Diatomaceous earth<br>White oil, 80 sec. visc. | 94 lbs.<br>6 pts. | Dust | 3 |
| *B* | 2 Klondike | Check, no treatment | ........ | ........ | ........ |
| XVII | (10 acres)<br>(Soil moisture control) | *No insecticidal treatment given* | | | |
| *A*[o] | 36 Eureka + 13 Neff | Moisture deficiency from Aug. 20 to end of season | | ........ | ........ |
| *B* | 42 Eureka + 7 Neff | | | ........ | ........ |
| *C* | 49 Eureka | Ample supply moisture throughout entire season | | ........ | ........ |
| *D* | 49 Eureka | | | ........ | ........ |
| | Totals—59 plots, 2,160 walnut trees, 1,048 peach trees, 125 acres | | | ........ | ........ |

[a] See pp. 529-530 for method of determining population rating.
[b] Data based on count of nuts on trees just prior to harvest (September 28).
[f] These were trees surrounding experimental plots for purpose of isolation, called "contact trees" in text.
[j] Actual increase in degree of infestation over check.
[m] Estimated average for all Eureka and Franquette trees in grove.
[n] Estimated average for all Klondike trees in grove.
[o] Below wilting point during late July.

TABLE 31—*(Concluded)*

| Number of applications | Date | Foliage injury "burn" | Average infestation, 1930 | | Population rating[a] | Results[b] | | | Per cent control |
|---|---|---|---|---|---|---|---|---|---|
| | | | Nuts per tree | Per cent | | Trees counted | Total nuts counted | Mean per cent infested | |
| 2 | 7/9, 8/1 | None | | | 1 | 20 | 4,824 | 13.9±1.6 | 15.3±21.9 |
| 2 | 7/9, 8/1 | Slight | .... | 10[m] | 1 | 14 | 2,994 | 3.5±0.7 | 78.7± 8.5 |
| .... | ............... | .................. | | | 2 | 19 | 3,016 | 16.4±3.5 | . .. |
| 2 | 7/15, 8/10 | None | 9 | .... | 2 | 16 | 1,855 | 11.1±1.3 | 40.0± 7 6 |
| 1 | 7/13 | None | 9 | .... | 1 | 16 | 1,493 | 24.5±1.8 | +30.0±15.1[j] |
| 1 | 7/13 | Slight | 22 | .... | 2 | 15 | 3,025 | 15.1±1.2 | 18.4± 7.6 |
| 1 | 7/12 | Slight | 8 | .... | 1 | 4 | 832 | 4.4±0.7 | 76.3± 7.6 |
| .... | ............... | .................. | 11 | .... | 2 | 15 | 2,779 | 18.5±1.6 | ............ . |
| 1 | 7/15 | None | .... | 10[n] | 1 | 3 | 1,050 | 2.8±1.5 | 10.0± 0.7 |
| .... | ............... | .................. | | | 1 | 2 | 837 | 3.1±0.7 | .......... . . . |
| .... | ............... | .................. | .... | 2 | 3+[p] | 4[q]<br>36[r] | 6,745<br>55,271 | 44.5±2.1<br>49.0 | 45[s] |
| .... | ............... | .................. | .... | 2 | 3+[p] | 4[q]<br>42[r] | 6,213<br>63,441 | 60.2±9.0<br>54.2 | |
| .... | ............... | .................. | .... | 6 | 3+[p] | 4[q]<br>49[r] | 2,745<br>37,895 | 89.9±0.9<br>95.1 | .... |
| .... | ............... | .................. | .... | 1 | 3+[p] | 48[r] | 23,636 | 96.4 | |
| .... | ............... | .................. | .... | .... | .... | 999 | 333,707 | .............. | .................. |

[p] Approximately 1400 flies colonized.
[q] Key trees.
[r] All trees in plot.
[s] Indicates effectiveness of soil moisture deficiency. Based on average of percentage infested on "key trees" plus "all trees" in plots *A* and *B* as compared with plots *C* and *D*.
[t] 2 per cent nicotine content.

*Plot Experiment I.*—Plots in this experiment total approximately 45 acres, which constitutes a portion of a 100-acre Eureka planting. Mature peach trees are interplanted between walnut trees in the rows from

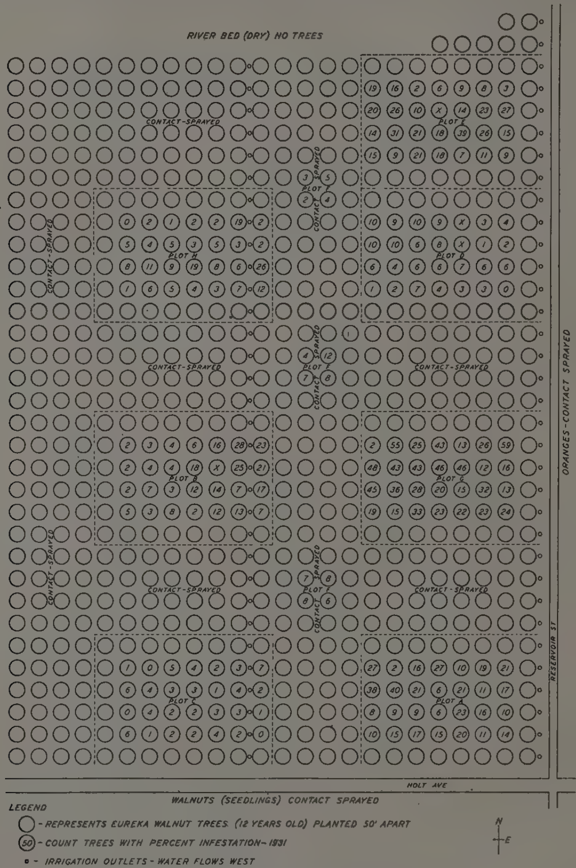



Fig. 72. Layout of plots in experiment I (45 acres); 1931 *Rhagoletis completa* control studies.

east to west. With the exception of plot E all plots were isolated in the same manner as in 1930. Figure 72 illustrates the arrangement of plots. Since the northern limits of the planting had been reached, it was not possible to maintain plots comparable in size and yet to isolate plot E

from plot D by the customary 4-row contact area. However, since the direction of the prevailing wind is from the northwest, and all trees in plot D as well as in the contact area to the west were sprayed, it is believed that there was not an appreciable drift of flies into plot E. In all instances, except in plot E, the peach interplants received the same treatment as the walnut trees.

Two dust applications of barium fluosilicate (plot C) afforded a fairly satisfactory degree of control. One dust application was approximately one-half as efficacious as two dust applications. The incorporation of 2 per cent fish oil in the dust mixture (plot B) materially increased the efficiency of one dust application (plot A). One spray application (plot D) resulted in a considerably higher degree of control than one dust treatment (plot A). In plot E, where the interplanted peach trees were not treated, the degree of control obtained was approximately one-half of that of plot D, in which the interplants were treated.

The results of one spray application in the contact area (plot F) were equivalent to those obtained in plot D, which received the same treatment. Only one tree separated the count trees of the contact areas from the nearest corner tree of a plot; the similarity of results in plots D and F might indicate that adequate isolation was provided under these conditions. However, it is doubtful if a buffer zone one tree deep between plots would preclude fairly free movement of flies from one plot to another. The method used in this experiment for plot isolation and the number and location of count trees to determine infestation were entirely satisfactory. Figure 72 illustrates the location of count trees in various plots and also furnishes a record of the percentage of infestation on individual trees.

The relation of fly population to walnut population, and the resulting percentage of infestation, is a matter of interest and one that requires thorough study. In plot H in 1930 there was an average of one infested nut per tree; while in 1931 there were approximately 25 infested nuts per tree. In plot G in 1930 the average of infested nuts per tree was 9; while in 1931 it increased to 78. The ratio of increase in infestation to expected female population in the two plots is not equivalent. Such factors as these are constantly operating in field control experiments; until detailed information regarding their nature is available, accurate comparisons of treated and untreated plots cannot be made.

*Plot Experiment II.*—In this grove, trees of the Placentia variety were planted alternately with Eurekas. The Placentia is very resistant to attacks by the fly, while the Eureka is very susceptible. This experiment was one of several intended to determine whether treating all trees within a solid block is necessary in order to obtain best results. The trees

were not very large; therefore 3 pounds of material per tree was a relatively heavy application. The data indicate that when all trees within a plot were treated, as good results were obtained from one application as were obtained from two applications. This evidence is contrary to that in experiment I. Where two applications were made (plot D) without treating the Placentia trees, as good results were obtained as in those plots where the Placentias were treated. Appreciable amounts of dust drifted into the adjoining trees, thereby enhancing the degree of control obtained for the plot. However, the results were poorest where one application was made without treating the Placentia trees. All treated plots had much lighter infestations than the untreated plots.

*Plot Experiment III.*—These tests were conducted in a grove of small Eureka trees that were interplanted with mature peach trees. They were designed to determine the value of treating peach interplants. The results do not show significant differences between the two treated plots, while the small percentage of infestation in both, when compared with the untreated plot, demonstrates the effectiveness of the material.

*Plot Experiment IV.*—This experiment was a direct comparison of one dust application of barium fluosilicate with one of basic lead arsenate. The data indicate that barium fluosilicate is superior to lead arsenate, even though the results in both instances were unsatisfactory.

*Plot Experiment V.*—In this experiment tests were conducted to determine whether or not the incorporation of an adhesive material in barium fluosilicate spray would result in increased effectiveness. Therefore light-pressed fish oil was included in the spray mixture. In comparing the degree of control obtained in this test with that of comparable plots similarly treated but without fish oil, no significant difference is apparent. The remaining plot in this experiment was dusted with hydrated lime. Results indicate that this material possesses considerable merit, thus warranting further trials. The nature of the action of this nonpoisonous substance is not understood.

*Plot Experiment VI.*—This is one of several experiments designed to determine how extensive an area surrounding individual trees of susceptible varieties must be treated in order to accomplish satisfactory control. The Eureka trees in this grove were interspersed singly among trees of the Placentia variety. Plot A consisted of three Eureka trees in different locations in the Placentia grove. These three trees were sprayed, while surrounding Placentia trees remained untreated. In plot B each Eureka tree was sprayed, together with a barrier zone of Placentia trees one tree deep, completely encircling the Eurekas. The data indicate unsatisfactory results in both plots, suggesting that the treated

barrier zone was not extensive enough. However, the infestation was lower in degree where the treated barrier zone was maintained.

*Plot Experiment VII.*—The purpose and layout of this experiment were similar to those of experiment VI but with synthetic cryolite in place of barium fluosilicate. The data corroborate experiment VI, in that a treated barrier zone one tree deep encircling individual susceptible trees is not extensive enough to give satisfactory results. If, as seems questionable, comparisons may be made between plots in different orchards, experiments VI and VII indicate that synthetic cryolite is superior to barium fluosilicate in effecting control of the fly.

*Plot Experiment VIII.*—This experiment was similar in purpose and layout to experiments VI and VII, but with a dust of natural cryolite as the treatment. The data indicate insignificant differences in the percentage of control obtained in treated and untreated plots.

*Plot Experiment IX.*—Plots A and B in this experiment afford a comparison of the effectiveness of natural cryolite with that of the synthetic product. Two dust applications of the natural product were more effective than one dust application of the synthetic material. However, the performance of one dust application of synthetic cryolite was considerably more effective than one dust application of barium fluosilicate in plot A of experiment I. The relative sizes of the trees in the two plots were such that the amounts of material applied per tree were comparable. Observations showed that synthetic cryolite adhered to the foliage better than did barium fluosilicate; this fact may partially explain any differences in percentage of control obtained with the two materials. The data furnished by plot C indicate that talc is apparently of no value in effecting control of the fly. Results in the contact area compared with those of contact areas in other experiments suggest that flies migrated into this area from plots C and E. The fly population in these two plots was relatively dense throughout the season.

*Plot Experiment X.*—This experiment did not yield any information regarding the efficacy of synthetic cryolite. The grove was heavily infested in 1930, but in 1931 only a very small percentage of flies emerged. Furthermore, the walnuts apparently were not in a susceptible condition during the activity of those flies that emerged.

*Plot Experiment XI.*—The efficacy of nicotine as a stomach poison in the control of the walnut husk fly warranted investigation. Experiment XI was one of several conducted to obtain information regarding this matter. Smith[35] determined that nicotine sulfate may be placed in solution in oil through the use of butanol as an intermediate solvent. The volatility of nicotine from a solution of this nature is greatly decreased and, therefore, when applied to walnut foliage the nicotine

offered a possibility of action as a stomach poison. However, the results of this test indicate that the material has no value in controlling the fly. A moderate degree of foliage injury was evident on all trees treated.

*Plot Experiment XII.*—The promising use of nicotine tannate in the control of the codling moth, *Carpocapsa pomonella* (Linn.)[21] suggested this test of the value of nicotine in the control of the walnut husk fly. Consequently two applications were made in plot A in comparison with two applications of barium fluosilicate in plot B. The size of the planting did not permit the usual isolation of plots. Each plot was only four trees wide, one adjoining the other. The results are not very significant, as evidenced by the percentage of control in the plots treated with barium fluosilicate when compared with the performance of this material in other experiments where only one spray application was made.

*Plot Experiment XIII.*—In this experiment the efficacy of nicotine sulfate plus sucrose as spray was compared with nicotine sulfate plus bentonite as a dust. In both mixtures the volatilization of nicotine was greatly retarded. The results indicate approximately equal degrees of control for these two materials. In both instances control was unsatisfactory.

*Plot Experiment XIV.*—When nicotine sulfate is incorporated with diatomaceous earth the volatilization of nicotine takes place very slowly. Therefore this mixture may be expected to possess value as a stomach poison. Experiment XIV served to compare the efficacy of nicotine sulfate plus diatomaceous earth as a dust with that of barium fluosilicate plus talc as a dust. The results indicate insignificant differences in percentage of infestation in the plot treated with nicotine and earth and the untreated check plot. The performance of barium fluosilicate plus talc was equivalent to that obtained in similarly treated plots in other experiments.

*Plot Experiment XV.*—This experiment was designed to determine the merits of finely ground tobacco dust containing 2 per cent nicotine, of diatomaceous earth, and of barium fluosilicate undiluted. The results indicate considerable value for tobacco dust, and none for diatomaceous earth. The results from a single application of straight barium fluosilicate are inconclusive, in view of the fact that there were more than twice as many infested nuts per tree in 1930 in this plot as in any other. A heavier infestation developed in treated plot B than in the untreated check plot. This illustrates the nature of some of the uncontrollable variables in experiments of this type.

*Plot Experiment XVI.*—In this experiment the diatomaceous earth and mineral oil dust was shown to be valueless in the control of the fly.

*Plot Experiment XVII.*—This was an experiment in applied ecology, in which an effort was made to manipulate the environment in such a manner as to render the walnuts resistant to oviposition attacks by the fly. The available information concerning irrigation and susceptibility

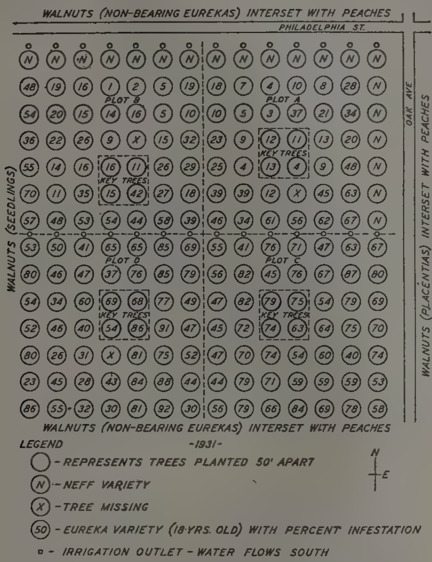



Fig. 73. Layout of plots in experiment XVII, on differential irrigation practice (10 acres), 1931 *Rhagoletis completa* control studies.

to infestation encouraged further investigation. Therefore the purpose of experiment XVII was to determine whether or not a relation exists between the amount of soil moisture present and susceptibility to attack by the fly and also to determine whether the degree of infestation can be influenced by manipulation of soil moisture. The portion of the experiment dealing with soil moisture was conducted in coöperation with O. L. Braucher.[5]

Four plots were laid out in a 16-year-old, 10-acre Eureka grove (fig. 73). A detailed history of the walnut-husk-fly infestation in this grove was available. The minimum number of trees in any plot was 36. Four

[5] Assistant in Orchard Management, University of California Citrus Experiment Station, and Field Investigator, California Walnut Growers' Association.

adjoining trees in the approximate center of each plot were selected for detailed observations and are referred to as "key trees" in the ensuing discussion.

The soil in plot A was permitted to go far below the "wilting point" during the latter part of July, which fact constituted the chief difference in soil-moisture manipulation between plots A and B. Both plots were subjected to a deficiency of soil moisture from the middle of August until after the crop was harvested. Therefore the moisture control exercised in these two plots was similar enough to warrant consideration of them as duplicates in this respect. Plots C and D were irrigated frequently enough to maintain soil moisture very close to the field capacity at all times. For this reason these two plots are considered as duplicates.

The fly population on the key trees was augmented in the early portion of the season by daily liberation of flies that were collected in the emergence cages. Equal numbers were liberated each day until a total of 348 flies had been colonized per key tree. From detailed studies of this grove during 1930, together with other data pertaining to pupal mortality, adult emergence, and percentage infestation, the approximate total number of flies present per tree was calculated. The average number of female flies per key tree, and the average number per tree in each plot (calculated from natural population, plus colonization) was as follows:

| Plot | Flies per key tree | Flies, per tree, entire plot |
|---|---|---|
| A | 196 | 41 |
| B | 211 | 54 |
| C | 306 | 146 |
| D | 199 | 40 |

Soil samples were taken at 10-day intervals, or more frequently when necessary, in the areas occupied by the key trees, and the moisture content was determined. Likewise at frequent intervals a composite sample of 50 walnuts was taken from the trees immediately surrounding the key trees in each plot, and the hardness of the husk was determined. These husk-hardness data are presented in figures 74, 75, and 76. This grove did not receive any insecticidal treatment for walnut-husk-fly control during 1931.

A double check on the degree of infestation was made. The first was a tree count prior to the beginning of harvest, to determine the percentage infestation at that time. These data for each tree are shown in figure 73. The second check was the harvest record in which the data for each key tree were kept separate; however, the data for the remainder of the trees in each plot were collected as a unit. A summary of certain of the

data obtained from this experiment, including the harvest record, is presented in table 31.

The data indicate a reduction in infestation of 45 per cent in plots A and B, when compared with plots C and D (table 31). There was a total of 118,712 nuts or possible host sites in the combined plots A and B and a calculated total of 3,744 females, or an average of 31.7 nuts per female. In the combined plots C and D there was a total of 61,531 nuts and a calculated total of 9,074 females, or an average of 6.8 nuts per female. In view of these data, the differences in percentage infestation between plots A and B, and C and D, as shown in table 31, cannot be considered significant. Therefore moisture control as practiced in these plots is apparently of negligible value in reducing infestation by the fly. Furthermore Braucher(7) concluded from his detailed study of soil variation within the confines of this 10-acre grove, together with the soil-moisture data, that the margin of safety to the general health of the tree and to the quality of the developing crop is not sufficient to warrant withholding moisture from the trees at such a critical time.

In comparing percentages of infestation from tree counts as shown in figure 73 with those for the harvest record which are given in table 31, certain features require clarification. The tree count to determine percentage infestation was made on September 26. At this time the husks of earlier-ripening walnuts were beginning to split. A tree count made 7 to 10 days later would not have yielded satisfactory data, since the husk of the uninfested walnut frequently darkens on the inside shortly after splitting, thereby rendering it somewhat similar in appearance to an infested walnut when viewed from a distance. A period of approximately 30 days usually elapses from the time the husks begin to split until the harvest is completed. Since characteristic husk blackness is the most feasible symptom to use in determining infestation from tree counts, those late-infested walnuts are not detected. However, the ratio of tree counts to final infestation was found to be similar in all plots.

This experiment supplied certain information regarding dispersion of flies within a grove. Percentages of infestation, as indicated by tree counts, are shown by the numerals within the tree circles in figure 73. These data indicate that the flies liberated on the key trees dispersed in all directions in a fairly uniform manner and reached the boundaries of the grove, a distance of 150 feet from the nearest key tree. It is the general opinion among many workers dealing with fruit flies, and particularly with economically important *Rhagoletis,* that the flies remain fairly well localized unless conditions exist that are unfavorable to oviposition. It is of interest to note the heterogeneity in degree of infestation in this grove and particularly within any one plot.

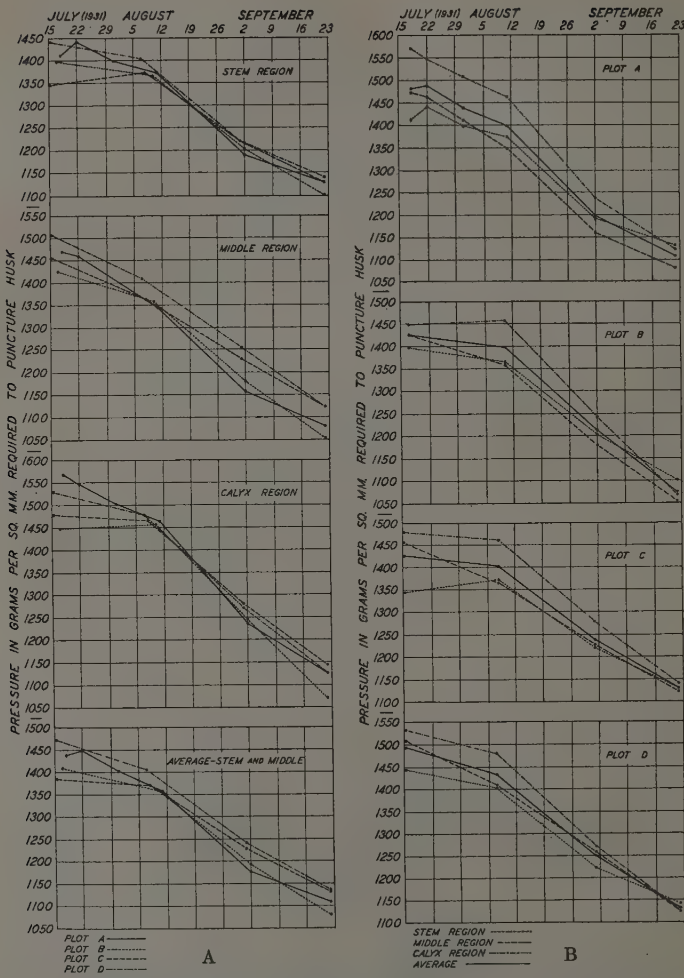



Fig. 74. Husk-hardness data regarding Eureka walnuts in experiment XVII, on differential irrigation practices, 1931 *Rhagoletis completa* control studies.

The husk-hardness data for the individual plots (fig. 74 *A*) show that in all instances the calyx region of the nut was hardest throughout most of the season of fly activity. There was no consistent significant difference in the hardness of the stem and middle regions in any of the four plots. A comparison of the husk hardness in the various regions in each of the four plots (fig. 74 *B*) does not present differences of sufficient

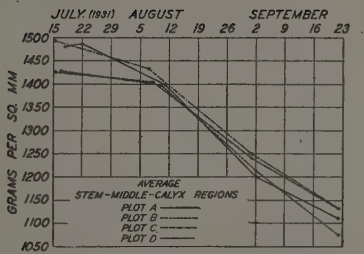



Fig. 75. Summary of husk-hardness data regarding Eureka walnuts in irrigation experiment XVII, on differential irrigation practices, 1931 *Rhagoletis completa* control studies.

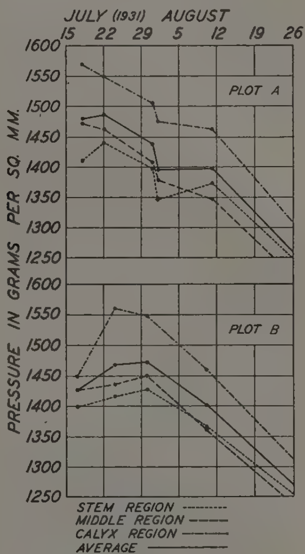



Fig. 76. Relation of irrigation to husk hardness in experiment XVII, on differential irrigation practices, 1931 *Rhagoletis completa* control studies. In plot A, the application of irrigation water on July 31 resulted in the husks' becoming softer immediately.

magnitude to be considered significant. The averages of the hardness of the three regions of the walnuts (fig. 75) in the four plots do not show significant differences between any of the plots.

Figure 76 shows that husk hardness is influenced to some extent by soil moisture under certain conditions. In plot A the soil moisture was well below the "wilting coefficient" on July 31. The average husk hardness of the three regions of walnuts at this time was approximately 1,445 grams per sq. mm. This plot was irrigated on July 31, August 1, and August 2, and the husk-hardness data were collected on August 2,

after irrigation was completed. At this time the average hardness of the three regions of the walnuts was approximately 1,395 grams per sq. mm. Therefore it is evident that the application of water to the soil was responsible for the husks' becoming 50 grams per sq. mm softer. Each region of the husk became softer, though the calyx region did not soften to the same degree as the stem and middle regions. Withholding moisture from the soil during July in plot A appears to have caused the husks to reach the peak of hardness by mid-July, at which time they began to soften; while in plot B, with normal irrigation during July, the husks reached the peak of hardness several weeks later.

*Discussion of Field Control Experiments in 1931.*—Seasonal conditions in 1931 were not considered entirely favorable for maximum economy in the biology of the walnut husk fly. Forty days intervened between the date when 50 per cent of the flies had emerged and that when the walnuts reached apparently optimum condition for oviposition. No doubt an appreciable number of flies died without depositing any eggs. Furthermore, the walnuts ripened approximately 10 days earlier than normally, thereby somewhat shortening the period for effective activity of the fly.

The size of the walnut crop was classified as "light." Therefore in most instances where ineffectual control was obtained, the degree of infestation was relatively heavy when expressed as a percentage.

The experimental plots supplied valuable information regarding the efficacy of various materials, the timing of treatments, and the method of application. In evaluating the results the probable error of the mean percentage of infested nuts is given, which indicates the significance of the data. The probable error of percentage of control in each instance indicates the reliability of the comparison of treated plots with untreated checks.

The following conclusions appear justifiable: Two dust applications of barium fluosilicate or cryolite, properly timed with respect to fly emergence, afforded satisfactory control. One dust application when the flies began to emerge was unsatisfactory; however, synthetic cryolite was apparently superior to barium fluosilicate under these conditions. One spray application of barium fluosilicate under these conditions was more effective than one dust application, though it did not afford satisfactory control. The incorporation of an adhesive material in the fluorine dusts is highly desirable. Of the various combinations employing nicotine as a stomach poison, nicotine tannate was the most effective; however, it was not so promising as either barium fluosilicate or cryolite.

All trees within a solid block must be treated to accomplish satisfactory results. Furthermore an area of trees or other vegetation adjoining susceptible varieties should be treated to insure maximum control efficiency. When a single tree or a few susceptible trees are growing interspersed with resistant varieties, treatment of a zone at least two trees deep is necessary for adequate protection. It is questionable whether or not the expense of this treatment would be justified by the increase in returns. Such a mixed planting condition is usually undesirable; therefore the removal of susceptible trees and replanting or topworking with resistant varieties is probably the best procedure. The available information regarding alteration of the environment through manipulation of soil moisture in an effort to inhibit oviposition activities of the fly is such that further experimentation is unwarranted.

*Adhesives Incorporated in Dust Mixtures.*—Fairly extensive field tests on both walnut and citrus trees have shown that in general the more commonly used fluorine compounds do not adhere well to foliage, but they adhere better when applied as spray than as dust. During August, 1931, an unseasonal rain of from 0.20 to 0.50 inch resulted in the loss of a large percentage of the barium fluosilicate and synthetic cryolite from the foliage of trees in dusted plots. Very appreciable amounts of those materials are lost from walnut foliage as a result of runoff in atmospheric dew. Since the cost of application of materials for walnut-husk-fly control favors the dust method, an effort was made to increase the adhesiveness of dust materials, particularly barium fluosilicate and synthetic cryolite. Hood[22] showed that the incorporation of fish oil in lead arsenate sprays greatly increased the period over which it adhered to the foliage of forest trees in the New England states. Marcovitch and Stanley[27] recommend fish oil in dusts of barium fluosilicate and cryolite used for the control of the Mexican bean beetle, *Epilachna corrupta* Muls., in Tennessee. Tests were first conducted here on citrus in 1929 using various percentages of a light-pressed herring oil and sperm oil incorporated in both barium fluosilicate and cryolite dust mixtures. Both talc and diatomaceous earth were used as diluents. Where 8 per cent of either of these oils was used, most of the material remained on the foliage through the winter rainy season.

For the tests with adhesive materials on walnuts during 1931, fish oil classified as light-pressed herring oil, raw linseed oil, cottonseed oil, and highly refined mineral oils of viscosities ranging from 60 to 100 seconds Saybolt were each used at concentrations by weight of 2, 4, and 8 per cent. Fiber talc and diatomaceous earth were each used as diluents with both barium fluosilicate and synthetic cryolite. In all instances where the oils were used at a concentration of 2 per cent the sticking

qualities of the dust were unsatisfactory; at 4 per cent the materials adhered fairly satisfactorily to the foliage. At a concentration of 8 per cent the adhesive qualities of the dust mixture were very good; however, where talc was used as the diluent the increased specific gravity of the mixture greatly reduced the mechanics of application of the dust. When diatomaceous earth was used as a diluent the incorporation of 8 per cent of any of the oils did not adversely affect the dusting qualities of the mixture; and in fact in one instance where 16 per cent of an 80-seconds-viscosity mineral oil was incorporated in the dust mixture of diatomaceous earth and barium fluosilicate, the dusting properties were not materially reduced. The experience gained in mixing these dust formulas corroborates the findings of Flint and Farrar,[14] that oil dusts cannot be satisfactorily mixed in the hopper of the regular self-mixing dusting machines. In the walnut-husk-fly control studies, the most satisfactory oil dusts were obtained where the oil was atomized into the toxic-diluent mixture in a special dust-mixing machine, and the final mixture broken up or fluffed by a rotary fiber brush revolving in a fine-mesh screen half-cylinder in the discharge mechanism of the mixer.

The field tests as conducted indicated the superiority of drying oil to mineral oil, at comparable concentrations, for sticking dust particles to foliage. The drying oil was completely dry within 2 to 3 days after application.

Diatomaceous earth was somewhat superior to other diluent materials tested, for it was not only especially light before the incorporation of oil, but also its dusting properties were not materially affected by the oil. However, in field application when mineral oils were incorporated as an adhesive agent in mixtures in which diatomaceous earth was the diluent, the rate of discharge of the dust could not be controlled as satisfactorily as when fish oil was used. Since the viscosity of the fish oil was nearly twice that of the heaviest mineral oils used, this factor may be related to the observed differences. When talc was used as the diluent, the rate of discharge could be controlled satisfactorily regardless of the type of oil that was incorporated for adhesive purposes.

Storage tests of fish-oil dusts show that the oil dries unless the air is excluded, thereby losing its effect as an adhesive. Furthermore, where higher percentages of fish oil (6 or 8 per cent) are used, particularly with talc as a diluent, if normal oxidation is permitted in storage, the dust mixture is likely to "heat up," thereby charring the material. Therefore the most satisfactory procedure, when practical, is to apply the fish oil-dust mixture soon after it has been mixed.

For the control of insects possessing sponging and sucking mouth parts, the probable performance of insecticide dusts in which oil had

been incorporated is a matter of conjecture. The use of dusts in which a sufficient amount of fish oil is included to insure maximum adhesiveness results in the particles' adhering very firmly to the leaf tissue; and the insect cannot "pick up" the particles of material as readily as when no adhesive material is used. When mineral oil is used in dust mixtures for adhesive purposes, the insecticide particles do not adhere to the leaf as firmly as when drying oil is used. On this basis, the mineral oil appears more promising than fish oil as an adhesive in dust mixtures for the control of the walnut husk fly.

Ferric oxide was used for adhesive purposes in several tests at concentrations of 20 and 50 per cent in the barium fluosilicate and talc mixture. The ferric oxide adherred to the foliage but there was apparently no appreciable increase in adherence of the insecticide.

## FIELD INVESTIGATIONS IN 1932

In 1932 there were 23 field control plots comprising approximately 55 acres. The experimental work on control was mainly to determine the efficacy of: (1) 20 and 30 per cent cryolite-dust mixtures, with mineral oil and with fish oil as the adhesive, when one application was made just prior to the peak of emergence in contrast to two applications at the previously mentioned times; and (2) several materials that had not been previously used on walnuts or in husk-fly control experiments. Most of the plots were located in groves that were used for experimental purposes in 1931; therefore the detailed history of infestation was available. Unfortunately it was not feasible to maintain untreated controls in any of the experiments; furthermore, practically every grove was treated in which an appreciable infestation was known to exist. Therefore an accurate evaluation of the performance of the materials and treatments was not possible except in comparison with one another. The conduct of the experiments was very similar to that of 1931. In two groves, the previously described system, that is, spraying of contact areas, was carried out in isolated plots. In the other experimental groves the plots were sufficiently large to insure satisfactory isolation of their central areas, from which the data were taken.

Adult emergence began June 29 and the seasonal peak was reached August 10 (fig. 45, p. 417).

The data obtained from the field control plots in 1932 are summarized in table 32.

*Plot Experiment I.*—This experiment was primarily designed to compare the efficacy of synthetic cryolite (sodium fluoaluminate), potassium fluoaluminate, magnesium arsenate, and acid lead arsenate. Cryolite and potassium fluoaluminate are apparently more effective in pro-

TABLE 32

Control Experiments in 1932

| Experiment | Number of trees and variety | Material | Concentration | Application: Method | Application: Amount per tree | Application: Number | Application: Date | Total cost per acre of walnuts (17 trees) | Per cent infested 1931 | Results[a]: Trees counted | Results[a]: Total nuts counted | Results[a]: Mean per cent infested |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| I | (10 acres) | | | | | | | | | | | |
| *A* | 25 Eureka | Cryolite[b]<br>Talc (fiber)<br>Mineral oil[c] | 20 lbs.<br>75 lbs.<br>5 lbs. | Dust | 3 lbs. | 2 | 7/29, 8/31 | $5.50 | 24.5 | 9 | 4,167 | 0.6± 0.2 |
| *B* | 25 Eureka | Potassium fluoaluminate<br>Talc (fiber)<br>Mineral oil | 20 lbs.<br>75 lbs.<br>5 lbs. | Dust | 3 lbs. | 2 | 7/29, 8/31 | 7.00 | 11.1 | 9 | 4,627 | 0.5± 0.2 |
| *C* | 25 Eureka | Magnesium arsenate<br>Lime (hydrated)<br>Mineral oil | 20 lbs.<br>75 lbs.<br>5 lbs. | Dust | 3 lbs. | 2 | 7/29, 8/31 | 9.00 | 18.5 | 9 | 6,067 | 1.9± 0.3 |
| *D* | 25 Eureka | Acid lead arsenate[d]<br>Lime (hydrated)<br>Mineral oil | 20 lbs.<br>75 lbs.<br>5 lbs. | Dust | 3 lbs. | 2 | 7/29, 8/31 | 6.00 | 15.1 | 9 | 6,107 | 2.9± 0.3 |
| *E* | 96 Eureka[e] | Cryolite<br>Fish oil[f] | 3 lbs.<br>1 pt.<br>100 gals. | Spray | 30 gals. | 1 | 7/27 | 5.25 | 4.4 | 9 | 4,409 | 0.5± 0.2 |
| *F* | 14 Eureka[g] | 7 trees treated as in Experiment *C*<br>7 trees treated as in Experiment *D* | ........ | Dust | 1½ lbs. | 2 | 7/29, 8/31 | ........ | 16.8 | 9 | 6,686 | 9.2± 1.4 |
| *G* | 1 Eureka[h] | Check, no treatment | ........ | ........ | ........ | .... | ........ | ........ | Approx. 15 0 | 1 | 435 | 21.8 |

[a] Data based on count of nuts on trees just prior to harvest (September 26).
[b] Synthetic cryolite used in all experiments.
[c] Mineral oil specifications: 95 sec. visc., 92 per cent unsulfonatable residue.
[d] Moderate foliage burn.
[e] These were trees surrounding experimental plots for purpose of isolation; called "contact trees" in the text.
[f] Fish oil classified as: light-pressed herring oil.
[g] Border row adjoining untreated grove; trees treated from one side only.
[h] Single tree in Placentia grove; 50 feet distant from Plot F.

TABLE 32—(*Continued*)

| Experiment | Number of trees and variety | Material | Concentration | Application: Method | Amount per tree | Number | Date | Total cost per acre of walnuts (17 trees) | Per cent infested 1931 | Results[a]: Trees counted | Total nuts counted | Mean per cent infested |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| II | (10 acres) | | | | | | | | | | | |
| *A* | 25 Eureka | Cryolite<br>Diatomaceous earth<br>Mineral oil | 20 lbs.<br>75 lbs.<br>5 lbs. | Dust | 3 lbs. | 2 | 7/29, 8/29 | $5.50 | 96 4 | 9 | 4,664 | 1.1± 0.2 |
| *B* | 25 Eureka | Cryolite<br>Diatomaceous earth<br>Fish oil | 20 lbs.<br>75 lbs.<br>5 lbs. | Dust | 3 lbs. | 2 | 7/29, 8/29 | 5.50 | 54 2 | 9 | 6,523 | 0.4±0.2 |
| *C* | 25 Eureka | Cryolite<br>Diatomaceous earth<br>Mineral oil | 30 lbs.<br>65 lbs.<br>5 lbs. | Dust | 3 lbs. | 2 | 7/29, 8/29 | 6.00 | 95.1 | 9 | 5,872 | 0.5± 0.2 |
| *D* | 25 Eureka | Cryolite<br>Diatomaceous earth<br>Fish oil | 30 lbs.<br>65 lbs.<br>5 lbs. | Dust | 3 lbs. | 2 | 7/29, 8/29 | 6.00 | 49.0 | 9 | 7,045 | 0.2± 0 2 |
| *E* | 96 Eureka[e] | Cryolite<br>Fish oil | 3 lbs.<br>1 pt.<br>100 gals. | Spray | 35 gals. | 1 | 7/27 | 6.00 | Approx. 75.0 | 9 | 8,761 | 0.8± 0.2 |

[a] Data based on count of nuts on trees just prior to harvest (September 26).
[e] These were trees surrounding experimental plots for purpose of isolation; called "contact trees" in the text.

TABLE 32—*(Continued)*

| Experiment | Number of trees and variety | Material | Concentration | Application: Method | Application: Amount per tree | Application: Number | Application: Date | Total cost per acre of walnuts (17 trees) | Per cent infested 1931 | Results[a]: Trees counted | Results[a]: Total nuts counted | Results[a]: Mean per cent infested |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| III | (15 acres)<br>Peach interplants | | | | | | | | | | | |
| *A* | 64 Eureka + 64 peach | Cryolite<br>Diatomaceous earth<br>Mineral oil | 20 lbs.<br>75 lbs.<br>5 lbs. | Dust | W[i] 2½ lbs.<br>P[i] ½ lb. | 1 | 8/11 | $2 75 | 6 2 | 12 | 7,669 | 0.4± 0.2 |
| *B* | 64 Eureka + 64 peach | Cryolite<br>Diatomaceous earth<br>Mineral oil | 20 lbs.<br>75 lbs.<br>5 lbs. | Dust | W 2½ lbs.<br>P ½ lb. | 2 | 7/28, 8/29 | 5 50 | 17.1 | 12 | 8,150 | 0.4± 0.2 |
| *C* | 64 Eureka + 64 peach | Cryolite<br>Diatomaceous earth<br>Mineral oil | 30 lbs.<br>65 lbs.<br>5 lbs. | Dust | W 2½ lbs.<br>P ½ lb. | 1 | 8/11 | 3.00 | 16.9 | 12 | 6,849 | 1.0± 0.2 |
| *D* | 64 Eureka + 64 peach | Cryolite<br>Diatomaceous earth<br>Mineral oil | 30 lbs.<br>65 lbs.<br>5 lbs. | Dust | W 2½ lbs.<br>P ½ lb. | 2 | 7/28, 8/29 | 6.00 | 30 7 | 12 | 7,179 | 0.6± 0.2 |
| IV | (10 acres) | | | | | | | | | | | |
| *A* | 61 Eureka + 61 Neff | Cryolite<br>Diatomaceous earth<br>Mineral oil | 30 lbs.<br>65 lbs.<br>5 lbs. | Dust | 3 lbs. | 1 | 8/12 | $3.00 | 12.8 | 16 | 9,021 | 2.8± 0.3 |
| *B* | 61 Eureka + 61 Neff | Cryolite<br>Diatomaceous earth<br>Mineral oil | 30 lbs.<br>65 lbs.<br>5 lbs. | Dust | 3 lbs. | 2 | 7/29, 8/27 | 6.00 | 14.5 | 16 | 9,661 | 1.0±0.2 |

[a] Data based on count of nuts on trees just prior to harvest (September 26).
[i] W=walnut; P=peach.

TABLE 32—(*Concluded*)

| Experiment | Number of trees and variety | Material | Concentration | Application: Method | Application: Amount per tree | Application: Number | Application: Date | Total cost per acre of walnuts (17 trees) | Per cent infested 1931 | Results[a]: Trees counted | Results[a]: Total nuts counted | Results[a]: Mean per cent infested |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| V | (9 acres) | | | | | | | | | | | |
| *A* | 10 Eureka[j] | Cryolite<br>Diatomaceous earth<br>Mineral oil | 30 lbs.<br>65 lbs.<br>5 lbs. | Dust | 3 lbs. | 2 | 7/28, 8/28 | $6.00 | Approx. 95.0 | 10 | 7,800 | 22.6± 2.2 |
| *B* | 10 Eureka[k] | Cryolite<br>Diatomaceous earth<br>Mineral oil | 30 lbs.<br>65 lbs.<br>5 lbs. | Dust | 3 lbs. | 2 | 7/28, 8/28 | 6.00 | Approx. 95.0 | 10 | 7,893 | 7.5± 0.9 |
| *C* | 130 Eureka[l] | Cryolite<br>Diatomaceous earth<br>Mineral oil | 30 lbs.<br>65 lbs.<br>5 lbs. | Dust | 3 lbs. | 2 | 7/28, 8/28 | 6.00 | Approx. 95.0 | 10 | 7,782 | 5.9± 0.4 |
| *D* | 6 Eureka[m] | Cryolite<br>Diatomaceous earth<br>Mineral oil | 30 lbs.<br>65 lbs.<br>5 lbs. | Dust | 3 lbs. | 2 | 7/28, 8/28 | 6.00 | Approx. 95.0 | 6 | 3,516 | 20.9± 1.6 |
| VI *A* | 5 Eureka[n] | Check, no treatment | ........ | ........ | ........ | .... | ........ | ........ | ........ | 5 | 1,288 | 57.2±11.5 |
| Totals—23 plots, 1054 walnut trees, 256 peach trees, 54 acres | | | | ........ | ........ | .... | ........ | ........ | ........ | 221 | 142,171 | ........ |

[a] Data based on count of nuts on trees just prior to harvest (September 26).
[j] West border row adjoining untreated Placentia grove 50 feet distant.
[k] Row adjoining plot *A*, 50 feet distant.
[l] Major portion of grove, exclusive of outer two border rows on east and west.
[m] East border row adjoining untreated Placentia grove.
[n] Untreated isolated susceptible trees in adjoining groves of resistant varieties.

ducing fly mortality than the two arsenicals tested. The data indicate that magnesium arsenate is more effective than acid lead arsenate; however, the difference is not great enough to be conclusive. Magnesium arsenate had not been used previously in these control studies; therefore the effect upon the tree was carefully observed. No injury resulted in this plot; however, in the acid lead arsenate plot moderate foliage burn was evident. This corroborates previous work with this arsenical, thus eliminating it from further consideration in this project.

One spray application of cryolite (plot E) is evidently as effective as two dust applications. (Other experiments in 1932 indicate that there is little difference in control between one and two applications of dust.)

The trees of plot F constituted a border row and were adjacent to an untreated Placentia grove at the regular planting distance of 50 feet. Furthermore this outer row of Eureka trees was only treated from one side, since the presence of a wire fence did not permit access to the other side. A strip of alfalfa was growing in the space between the Eureka row and the first Placentia row. The control was unsatisfactory, apparently either because the trees were sprayed only from one side or because the adjoining trees and alfalfa were not treated. Flies were commonly observed on the Placentia trees throughout the season.

Plot G consisted of a single untreated Eureka tree that was growing in the row of Placentia trees adjoining plot F. Flies became very abundant on this tree as the season progressed. While this single-tree plot does not constitute a representative control, the data, together with the data from plot F, are strongly indicative of what would probably have occurred in the other plots had not action of the applied materials been effective.

*Plot Experiment II.*—The cost of cryolite control warranted investigation of the comparative efficacy of 20 per cent and 30 per cent cryolite dust mixtures, each with fish oil and mineral oil incorporated. Accordingly plots were treated in a manner designed to show the merits of each combination.

Fourteen emergence cages were located in this grove and a daily record of fly emergence was kept. As a result of the relatively heavy infestation here in 1931, enormous numbers of flies emerged during the 1932 season.

Data regarding the hardness of walnut husks, collected from this grove at intervals throughout the season, showed that the walnuts were in susceptible condition for infestation throughout the period of adult emergence. Therefore it seems logical to conclude that the status of infestation in the grove at the end of the season was the direct result of the treatments given.

The very light infestation in all plots indicates that the materials used were highly efficacious in controlling the fly. The existing differences in degree of infestation in the various plots are not of sufficient magnitude to warrant conclusions as to the comparative value of the individual combinations.

One application of cryolite as spray (plot E) is apparently as effective as two dust applications.

*Plot Experiment III.*—The 1931 experiments demonstrated that one dust treatment applied at the beginning of fly emergence produced unsatisfactory results. Yearly observations regarding the peak of oviposition have shown that most of the eggs are deposited during the latter half of August, regardless of when the peak of fly emergence is reached. Therefore it was important to compare the effectiveness of one treatment applied before August 15 with two treatments timed as recommended previously. Further information regarding the comparative values for 20 per cent and 30 per cent cryolite dust was also desirable.

The data of experiment III indicate an insignificant difference in efficacy between the two concentrations tested, and between one and two applications. Apparently one application of 20 per cent cryolite dust properly timed produces satisfactory results.

*Plot Experiment IV.*—This experiment was outlined to obtain further information regarding the efficacy of one and of two applications of 30 per cent cryolite dust timed as described in experiment III. The data indicate a difference of approximately 2 per cent in degree of infestation in favor of two applications, but this difference does not appear to justify the extra expense of two treatments.

*Plot Experiment V.*—This experiment furnished additional information regarding the value of treating a bordering zone of nonsusceptible trees in order to obtain satisfactory control. The infestation in this grove in 1931 was uniformly heavy. Treatment in 1932 was according to recommendations. This Eureka grove was bounded by untreated Placentia groves on the east and west, by a paved street on the north, and by a grove of small Payne trees, which were treated, on the south. This Eureka grove, and the Placentia groves adjacent to the east and west, were very similar in size of trees and in soil and tree management. Flies were commonly observed throughout the season on the adjacent west row of Placentia trees. Figure 77 shows the location of the several plots.

In plot A, the west border row of Eureka trees adjoining the untreated Placentia grove, a 23 per cent infestation developed, while in plot B (which was the next row east) the infestation was 8 per cent, and in plot C, in the center of the grove, 6 per cent. In plot D, the east border

row adjacent to the untreated Placentia grove, the infestation was 21 per cent.

The data demonstrate conclusively that failure to treat an adjoining zone of at least one row of resistant trees materially affected the degree of control obtained in the border row of this Eureka grove.

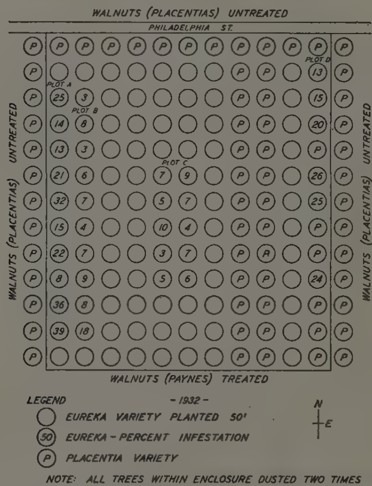



Fig. 77. Layout of plots in experiment V (9 acres), which demonstrates the necessity of proper isolation through treated barrier zones; 1932 *Rhagoletis completa* control studies.

*Plot Experiment VI.*—Observations were made on a few untreated Eureka trees singly isolated in groves of nonsusceptible varieties from various locations in the infested area. The infestation varied from 22 per cent to 99 per cent. Definite information regarding the degree of infestation in 1931 is lacking; however, the trees were reported to have been moderately to heavily infested. While these data cannot be considered as a control, they are indicative of the potential infestation in the experimental plots, which probably would have developed had no control measures been applied.

*Discussion of Field Control Experiments in 1932.*—The size of the 1932 crop of walnuts was considered large; therefore the performance of materials expressed in terms of percentage infestation is not directly comparable to that of the 1931 season, when the crop was small. How-

ever, size of crop in the experimental groves, with the exception of experiment V in 1931, was sufficiently uniform both seasons to warrant relative comparison of results in the various plots. The size of crop in experiment V in 1931 was larger than average for that year; thus with the high percentage (95 per cent) infestation the fly population in 1932 was considerably larger than in other groves.

Several important factors were responsible for mitigating the degree of infestation this season. Emergence records show that only approximately 67 per cent of the 1931 pupae produced annual-generation flies. Similar limiting effects were also operative on the emergence of biennial-generation flies. The walnuts ripened fully three weeks earlier than normal, thereby shortening the oviposition period somewhat. Furthermore, an appreciable percentage of those nuts in which oviposition took place after September 15 were not injured nor did the larvae mature. Data collected during harvest indicate that approximately 25 per cent of the infested walnuts ripened before the larvae attained sufficient size to cause the husks to blacken as a result of their feeding. Thus the degree of actual damage was appreciably lessened. To partially compensate for these factors adverse to the insect, the walnut husks reached the susceptible stage for oviposition at a more nearly optimum time for the fly with respect to the seasonal peak of emergence. The field-count method evidently indicated that under the circumstances existing this season, the mean percentage infestation more closely approached the degree of actual injury than it did in 1931. The data are believed to be as nearly comparable as conditions permit with this type of field-plot experimentation.

From the foregoing facts and in consideration of the data obtained from the field control plots, it seems logical to conclude that: One properly timed treatment of synthetic cryolite applied as dust or spray will afford satisfactory control. Two dust treatments of either potassium fluoaluminate or magnesium arsenate are also highly efficacious. Acid lead arsenate is an effective material but is unsafe to use from the point of view of tree health. Mineral oil and fish oil are apparently of equal value as an adhesive in the dust mixtures used. To insure satisfactory protection of susceptible varieties through insecticidal treatment, a zone of adjacent vegetation from 50 to 100 feet wide must be similarly treated.

## RECOMMENDATIONS TO GROWERS ON CONTROL OF THE WALNUT HUSK FLY

On the basis of field control experiments, either synthetic cryolite[6] or barium fluosilicate is recommended for general use in the control campaign in the infested area. Synthetic cryolite from different manufacturers has been found to vary in the content of sodium fluoaluminate, uncombined sodium fluoride, and various sulfates, and also in solubility in water and such physical properties as fineness and bulk or weight. Therefore the following tentative specifications are suggested:

Sodium fluoaluminate ($Na_3AlF_6$): not less than 97 per cent
No uncombined sodium fluoride
Free of sulfates
Solubility: not more than 1 gram in 1,600 cc water at 20° C
Mesh: 100 per cent through 200 mesh per inch; not less than 75 per cent through 325 mesh per inch (U.S. Bur. Standards Sieve Series No. 325)
Volume (screened loose): not less than 48 cubic inches per pound

An adhesive is necessary in both sprays and dusts when either cryolite or barium fluosilicate is used. After preliminary experiments with various types of fish oils and vegetable oils, and with highly refined mineral oils of different viscosities, a mineral oil of the following specification appears most promising as an adhesive:

Viscosity: 95 seconds Saybolt
Sulfonation: 90 per cent

As a spray the recommended formula is:

| | |
|---|---|
| Synthetic cryolite or barium fluosilicate | 3 pounds |
| Mineral oil | 1 pint |
| Water | 100 gallons |

From 30 to 40 gallons per average-sized tree affords satisfactory coverage.

The dust method of treatment is less expensive than the spray method. The recommended formula is:

| | |
|---|---|
| Synthetic cryolite or barium fluosilicate | 30 per cent |
| Diatomaceous earth | 65 per cent |
| Mineral oil | 5 per cent |

From 3 to 4 pounds per average-sized tree affords satisfactory coverage.

[6] Towards the termination of this project and afterwards, the preparation of natural cryolite for insecticidal purposes has been greatly improved and field trials elsewhere on other insects have demonstrated its efficacy in comparison with other fluorine compounds. Therefore it appears that this material may also be satisfactorily employed in the control of the walnut husk fly.

The proper diluent material for the dust mixture is of considerable importance. Of the many noncalcium materials tested, a special grade of diatomaceous earth and fiber talc appears most promising. Where an adhesive consisting of either mineral oil or fish oil is required in quantities of from 5 to 10 per cent, the diatomaceous earth is most satisfactory. Without an adhesive, fiber talc is entirely satisfactory. Since in this instance an oil adhesive is required, diatomaceous earth of the following tentative specifications is suggested:

Silica as diatomes: not less than 90 per cent
Free of calcium
Moisture: not more than 5 per cent
Mesh: 100 per cent through 200 mesh per inch; not less than 75 per cent through 325 mesh per inch (U.S. Bur. Standards Sieve Series No. 325)
Volume (screened loose): not more than 190 cubic inches per pound.

When this type of diluent is used, the bulkiness of the mixed dust possesses certain advantages, particularly from the point of view of application. The operator may permit a large volume of dust to discharge continuously while treating an individual tree and not be so likely to apply more than the relatively small dosage of 3 pounds specified. Talc is relatively heavy and therefore the operator must regulate the discharge carefully in order to prevent the application of a greater number of pounds per tree than is required. Because of weather conditions, practically all dust treatment is applied during the night; therefore the application is likely to be more uniform with a bulky dusting material. The disadvantages of this type of material are that the mixing costs are increased slightly, more sacks or containers are required, and stops for filling the dusting machine must be more frequent.

Proper mixing of the ingredients in the dust mixture is essential to insure best results. A modern machine designed for mixing of dust insecticides should be employed. Analyses have shown that a minimum time of 5 minutes is necessary to properly mix cryolite or barium fluosilicate and diatomaceous earth. After the oil is atomized into the fluoride-earth mixture, an additional 10 minutes' mixing is necessary to mix the oil thoroughly with the dust. Thus a total running time of 15 minutes per batch is required.

To insure adequate protection within a grove through insecticidal treatment, a zone of adjacent vegetation from 50 to 100 feet wide must be treated simultaneously with the infested grove. In view of the limited duration of the experimental work with these materials, two treatments are recommended, the first when adult emergence becomes regular, and the second approximately 4 weeks after the first. The 1932 data indicate that probably one properly timed application will afford satisfactory re-

sults. However, more extensive observations regarding seasonal history and seasonal host resistance are necessary before definite conclusions can be made regarding the most economical control measures.

## SUMMARY

*History.*—*Rhagoletis completa* Cresson was introduced into California prior to 1926 from the central region of the United States (about the 100th meridian), where it is apparently indigenous. It was probably introduced as larvae or pupae in black walnuts. In 1927 the species assumed major economic importance as a pest of certain commercial varieties of Persian walnut, *Juglans regia.* The area of infestation has increased yearly, and in 1932 it comprised approximately 500 square miles and included over 2,000 acres of commercial varieties of Persian walnut. Taxonomists confused this insect with *R. juglandis,* and it was not until 1929 that it was found to be undescribed. Cresson then considered it a subspecies of *R. suavis.*

*Taxonomy and Technical Description of Stages.*—Studies dealing with large series of specimens of *Rhagoletis suavis* and its subspecies *completa* from different areas resulted in the elevation of *completa* to specific rank. Characteristic differences are evident in the pattern of the infuscated areas of the wing and in male genitalia. *R. completa* is of a general tawny color with yellowish-white markings. The wings are hyaline with three parallel transverse infuscated bands. The distal band continues along the anterior margin of the wing to the apex. Length, 4 to 8 mm.

The egg of *Rhagoletis completa* is somewhat curved in shape and pearly white in color. First-instar larvae are semitransparent, and are characterized by the absence of anterior spiracles, the presence of two peritremes on each posterior spiracle, and a tooth on the blade of each oral hook. Second-instar larvae possess anterior spiracles and three peritremes on each posterior spiracle. Mature third-instar larvae are creamy white. The tooth on the oral hook is absent. There are 14 tubercles situated on the posterior body segment. These structures, together with the angle that the lower peritreme makes with the horizontal, serve to identify the species. The pupa is somewhat barrel-shaped and of straw color.

*Related Species Attacking Walnuts.*—*Rhagoletis suavis,* the walnut husk maggot, occurs throughout most of the eastern United States on the black walnut, *Juglans nigra.* This species is reported to be economically important as a pest of Persian walnut in New York, Pennsylvania, and Maryland. *R. juglandis* is probably a Mexican species and is recorded from southern Arizona and Chihuahua, Mexico. Serious infesta-

tions have been observed on Persian walnut in Arizona, where it also attacks the native black walnut, *J. rupestris. R. boycei* is recorded from southern Arizona. It probably attacks wild and cultivated walnuts. Adults of these three species are illustrated.

*Common Name.*—"Walnut husk fly" was proposed as the common name for this insect in 1929, and was formally accepted in 1930. At that time the species was believed to be *Rhagoletis juglandis.* Since this common name was actually intended for this particular insect, it is retained, although *R. completa* is now known to be distinct from *R. juglandis.*

*Distribution.*—Authentic records show that *Rhagoletis completa* occurs in Nebraska, Kansas, Oklahoma, Texas, New Mexico, and California.

*Host Studies.*—The walnut is the preferred host of this insect, though infestations of minor importance have been observed in the peach under field conditions. Under laboratory conditions several larvae reached maturity in the tomato, and pupated. The late-maturing thick-husked varieties of walnut are most susceptible to attack. The nature of this very marked varietal susceptibility is apparently related to the hardness of the husk of the different varieties at the time of oviposition. The green husk tissue becomes harder as the walnut develops, reaching a peak of hardness usually during late June or early July, and then softening during late August and early September. Females are unable to puncture the husk for egg deposition at the peak of hardness; therefore oviposition takes place on the descending slope of the seasonal-husk-hardness curve. The husk of the so-called "resistant" varieties is generally not soft enough for egg deposition until several weeks prior to maturity of the nut and subsequent harvesting. Extensive data regarding husk hardness over a period of four years were obtained through the use of a modified Jolly balance.

Laboratory studies of possible hosts showed that females attempted oviposition in all fruits and tubers placed in cages with them. Eggs were deposited below the surface of the skin in tangerine, Mediterranean Sweet orange, apple, pear, quince, plum, prickly pear, potato, eggplant, and bell pepper. Larval maturity was not reached in any instance. Females vigorously attempted to insert eggs below the skin surface of the ripe Valencia orange and grapefruit. The texture of the lower skin tissue prevented them from making a cavity; however, the ovipositor was readily inserted into the tissue. Usually a single egg was placed into the small, shallow hole made by the ovipositor and other eggs were voided on the surface nearby. In all cases the eggs dried out before hatching.

Eggs were artificially placed in the pulp tissue of oranges, and larvae hatched and reached maturity within the fruit.

*Injury and Economic Importance.*—The principal type of injury results from the feeding of larvae within the green husk, thereby causing internal decay which permanently blackens the shell of the walnut. Such affected walnuts become "culls" with a resultant loss in value to the producer of approximately 50 per cent. The secondary type of injury is manifested by a reduction in quality of the kernels of infested nuts. The net loss in value varies from 0 to 25 per cent, according to seasonal conditions. Infested walnuts generally become "sticktights," resulting in increased harvesting costs. Other economic considerations are the costs incident to enforcement of regulatory measures for the prevention of artificial spread into uninfested areas.

*Adult.*—Methods were developed whereby various laboratory experiments involving thousands of adults were fairly satisfactorily conducted. An inverted battery jar, resting on plate glass and with wire-screen vent at the base, produced very favorable humidity conditions for longevity. Liquid food consisting of sucrose or honey was adsorbed in cotton and placed in the cages in one-half of a small petri dish.

Detailed records of adult emergence from the soil over a five-year period, involving over 37,000 flies, show that the seasonal peak varies considerably. Winter temperatures are apparently of prime importance in this connection. For the five-year period an average of 71 per cent of the flies emerged the year after pupation and are classed as annual-generation individuals; 29 per cent emerged the second year after pupation and are classed as biennial-generation individuals; while a few flies did not emerge until the third and fourth years after pupation and are classed as multi-annual-generation individuals. The daily rate of emergence within a single season is apparently influenced by daily mean temperature. High temperature is accompanied by high rate of emergence and low temperature by low rate of emergence. When the former condition exists, the major portion of those individuals that emerge during one day do so in the forenoon before the temperature reaches the daily peak.

Adults are attracted to a slight degree by the products of fermenting molasses. Outdoor laboratory tests, wherein over 100 organic chemicals were used in chemotropic studies, gave neutral results. Simple phototropic tests produced inconclusive data. Flies are apparently neutral in their reaction to anemotropic and thermotropic stimulation. They show strong negative geotropism immediately after emerging from puparia.

Honeydew, resulting from infestation by aphids, spores of yeast and fungi occurring naturally on the trees, and moisture, mainly in the form

of atmospheric dew, appear to constitute the food of the adult. Under field conditions the flies generally feed most actively in the morning hours after the sun rises and until the dew has completely evaporated. Laboratory studies demonstrated that the flies ingest very small, solid particles of matter.

Flies have been observed to travel on the wing from one tree to another. Other evidence regarding flight and dispersion is circumstantial: when food and oviposition conditions are favorable, the flies apparently remain localized on individual or closely adjacent trees; however, when either of these factors is adverse, migration is evidently stimulated.

Sucrose applied under uniform conditions to walnut foliage served to congregate a portion of the flies present, and thereby enabled a comparison of the relative density of the fly population on individual trees. The data show that the movement of flies on trees is closely related to sunshine, temperature, and humidity.

The average length of life of adults under laboratory conditions was approximately 40 days, and the most aged individuals lived 85 days. Under field conditions the average length of life is probably from 30 to 40 days. Without food, the greater portion of the flies died within 50 hours. When daily peak temperatures ranged from 95° to 100° F, the length of life was materially shortened; however, under optimum humidity conditions flies were not killed when the temperature remained at 114° F for several hours. High relative humidity is apparently a very important factor in longevity.

From preliminary nutritional studies dealing with several proteins, carbohydrates, and minerals, the following indicative information was obtained.

Proteins: Apparently yeast decreased longevity and fecundity; glycocoll decreased longevity though it increased fecundity; and both urea and ammonia increased longevity and fecundity.

Carbohydrates: Apparently sucrose, levulose, dextrose, and honey were essential for longevity and fecundity; both honey and levulose reduced longevity slightly in comparison with sucrose, though fecundity was increased; dextrose reduced longevity in comparison with sucrose, without affecting fecundity; and dextrin had little or no food value.

Minerals: Apparently zinc and copper in the diet increased longevity and fecundity.

pH of media: Apparently no relation exists between food of pH from 3.8 to 9.5, and longevity and fecundity.

The most important male genital structures are illustrated. They are: the testes, seminal vesicles, vasa deferentia, accessory glands, ejaculatory duct, seminal pump, aedeagus, and claspers.

The most important female genital structures are illustrated. They are: the ovaries, oviducts, vagina, spermathecae, accessory glands, ovipositor sheath, and ovipositor.

Copulation first took place 6 or 8 days after the adults emerged from the soil. Under natural conditions it was most commonly observed during late afternoon, and usually immediately followed oviposition. The indications are that it was of frequent occurrence.

The first eggs in the ovaries were completely developed in from 10 to 20 days after the female emerged from the soil. This preoviposition period in a large number of individuals averaged 18 days under laboratory conditions.

It is estimated that under optimum field conditions females deposited from 200 to 400 eggs. Fecundity was greatly reduced in the laboratory and the maximum number of eggs deposited by one female was 84, in a total of 7 cavities. In oviposition, a cavity just below the surface of the husk is produced by the female, the ovipositor being used to lacerate the inner tissue. Eggs are deposited in batches of approximately 15 and usually all eggs within an individual cavity are deposited by one female at one insertion of the ovipositor. Eggs are deposited in healthy tissue only. Data for a four-year period show that 72 per cent of all cavities of eggs were located in the stem region of the husk, 24 per cent in the middle region, and 4 per cent in the calyx region. Females show a slight preference for the nuts in the middle and upper portions of the tree for oviposition, rather than in the lower portion.

*Egg.*—The average length of incubation period under field conditions was 120 hours. It was 72 hours under laboratory conditions. Egg mortality in the field was approximately 20 per cent and was mainly due to infertility and the work of natural enemies.

*Larva.*—Newly hatched larvae remained alive without food for 6 to 12 hours. The larvae consume only healthy tissue for food. They have gregarious tendencies in feeding. The average length of the instars under field conditions was: first instar, 9.7 days; second instar, 13.0 days; third instar, 14.1 days; and total development, 36.8 days. During the early portion of the season a number of larvae reached maturity in 18 or 20 days. Larval mortality within the walnut husk approximated 25 per cent.

When maturity was reached, the larvae issued from the husk and dropped to the soil, where they burrowed downward and later pupated. The greater percentage of daily emergence of larvae from the walnuts occurred between the hours of 5:30 and 8:00 a.m., which fact indicates a relation with temperature. The larvae show marked positive geotropism, and apparently react negatively to light. The depth that larvae

penetrated the soil to pupate varied from ½ to 7 inches, according to the soil type, degree of moisture present, and the existing state of cultivation.

Limited laboratory studies regarding the effect of temperature on larvae showed that at 30° F for 35 hours the mortality was fairly high; however, when removed and placed at 72° F, pupation was materially stimulated by the exposure to low temperature. An exposure of ½ hour at 115° F proved fatal to a high percentage, while exposure for ¾ hour resulted in total mortality.

*Pupa.*—The puparium was completely formed within 24 hours after the larva entered the soil. An additional larval molt occurred within 36 hours after the formation of the puparium. The true pupal stage was reached within 145 to 175 hours after the larva entered the soil. Normal summer temperatures of the soil in total sunlight proved fatal to pupae located in the upper portion of the soil; however, most of the pupae were buried to a depth of from 3 to 8 inches by the usual cultivation practices. Pupal mortality was variable, though considerable, in all instances recorded.

Data obtained regarding dormancy indicate that a fairly definite amount of heat units supplied by a range of fluctuating temperatures are important in the termination of this condition. Tests of various chemicals showed that both potassium thiocyanate and thiourea apparently exerted a slight effect upon the termination of dormancy.

*Seasonal History.*—The data show that accumulated soil-temperature conditions during dormancy and host resistance exert a profound effect upon seasonal activity of the fly. The relation of winter temperatures to time of seasonal emergence has already been pointed out.

Oviposition data show that the peak of egg laying each year for the five-year period was reached between August 29 and September 5, despite wide variation in the seasonal median of adult emergence. Evidently hardness of the green husk is the most important physical factor relating to time of oviposition as well as to varietal susceptibility.

The earliness of walnut harvest influences larval emergence from the husks. Under average conditions approximately 75 per cent have issued at the time of harvest; however, in 1932 harvest was about 20 days earlier than normal and only 24 per cent had emerged at that time.

*Natural Enemies.*—The walnut husk fly is remarkably free from important natural enemies. Several species of parasitic fungi have been cultured from dead adults. The mite, *Pediculoides ventricosus* New., and the anthocorid, *Triphleps insidiosus* (Say), prey upon the eggs. Several other species of common predators, including spiders, a reduviid, a chrysopid, and ants, feed upon the larvae, pupae, or adults. Two

general feeding parasites of dipterous pupae, the chalcid, *Spalangia rugosicollis* Ash., and the proctotrupid, *Galesus* sp. near *atricornis* Ash., have been reared from *Rhagoletis completa* at Manhattan, Kansas. The opiine larval parasites, *Opius humilis* Silv. and *Diachasma tryoni* Cam., have been introduced from Hawaii, and the former was recovered in the field in 1932.

*Scavenger Species Inhabiting Decaying Walnut Husks.*—More than 30 species of scavenger insects, mainly Diptera, have been reared from decaying walnut husks. The flies, *Euxesta putricola* Cole, *Lonchaea occidentalis* Mall., *Muscina assimilis* Fall, *Fannia canicularis* (Linn.), and *Drosophila* spp., are most commonly observed. All scavengers are of negligible economic importance.

*Laboratory Toxicological Investigations.*—A satisfactory method was devised whereby a comparison of the relative speeds of toxic action of various materials upon adult flies was possible. Tests of this nature were conducted during 1929, 1930, 1931, and 1932. The results of these tests are graphically presented. Of the arsenicals tested, basic lead arsenate was consistently slowest in speed of lethal action, though aside from magnesium arsenate it was the only one of the group that did not cause foliage injury. With respect to magnesium arsenate, one season's observations indicate that it is safe to use on walnut foliage; however, its lethal action is apparently not appreciably more rapid than that of basic lead arsenate. Of the fluorine compounds tested, synthetic cryolite (sodium fluoaluminate), barium fluosilicate, and potassium fluoaluminate are the most promising from the point of view of toxicity to the insect and tree tolerance. The copper compounds tested offer promise with respect to lethal action, though they are deleterious to walnut foliage. The nicotine compounds and combinations tested are effective as stomach poisons on the flies, though they do not appear to be as practicable in the field as the several fluorines mentioned. Of the inert diluents, either diatomaceous earth or talc is satisfactory. Hydrated lime employed as a diluent with synthetic cryolite materially retarded lethal action. All diluent materials tested, whether chemically active or inert, were lethal to the flies. The nature of their action is unknown; however, hypotheses are suggested for the mode of action of several of these materials. The incorporation of either mineral oil, vegetable oil, or fish oil at a concentration of 5 per cent for adhesive purposes, did not retard lethal action of the insecticide dust mixture. However, at a concentration of 12 per cent, both mineral oil and fish oil apparently inhibited the speed of toxic action.

*Field Investigations.*—The field investigations dealt mainly with plot experiments to determine the efficacy of various materials, concentra-

tions, and methods and time of application. Wherever possible, plots were isolated from each other and from adjoining properties to preclude the undue influence of uncontrollable variable factors. The application in all plots was very thorough and the infestation data were analyzed with the aid of simple statistical methods. In many instances erratic and inconclusive data were obtained. A summary of the more important results of the field experiments follows:

Available information indicates that control of the fly by altering the environment through manipulation of soil moisture in an effort to inhibit oviposition activities is impractical.

The application of basic lead arsenate does not afford a satisfactory means of controlling the fly though it is generally most effective when applied two or three times as a spray, and when thorough coverage of the trees is obtained. Synthetic cryolite and barium fluosilicate applied as a spray or dust are satisfactory insecticides for controlling this insect.

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